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# Estimation of the Number of Days Required to Determine Usual Antioxidant Intakes and Assessment of the Prevalence of Nutrient Inadequacy Among College Students

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## Recommended Citation

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Estimation of the Number of Days Required to Determine Usual  
Antioxidant Intakes and Assessment of the Prevalence of Nutrient  
Inadequacy Among College Students

Catherine Davis

B.S., University of Georgia, 2009

A Thesis

Submitted in Partial Fulfillment of the

Requirements for the Degree of

Master of Science

at the

University of Connecticut

2011

APPROVAL PAGE

Master of Sciences Thesis

Estimation of the Number of Days Required to Determine Usual  
Antioxidant Intakes and Assessment of the Prevalence of Nutrient  
Inadequacy Among College Students

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2011

## **Acknowledgements**

First and foremost I would like to thank God for his continual grace, for without him, I would not be who I am today.

I would like to thank my major advisor Dr. Chun. Without your continual encouragement, guidance, and standards of excellence in our lab, I would not have been able to accomplish all that I have during my time at UCONN. You have an admirable work ethic and are always considering our future in anything you ask us to do. I have thoroughly enjoyed working with and I thank you for every opportunity you have provided me.

I would also like to thank my committee members Dr. Fernandez and Dr. Rodriguez for their guidance over the past two years on my thesis as well as in the classroom. I have learned so much from the both of you through your example of hard work and diligence. I greatly appreciate your help with my thesis and manuscripts.

I would like to thank my labmates, Meng, Ying, and Sang Gil. I really cannot thank you enough. Thank you for your daily encouragement, for your support and for your friendship. I have loved working with you in all our studies.

Thank you to the faculty and staff here in the Nutritional Sciences Department for your guidance and education inside the classroom and out. I would also like to thank Dr. Chen and Vickie Zhang from the Department of Statistics at UCONN for their help with the statistical analysis of this project. I would like to thank the USDA Hatch for the funding of this study and all the study participants for their diligence to finish all those dietary records.

I would like to thank the very special people in this graduate department who have become friends as well as my friends back home for their support and encouragement. I would especially like to thank Mariana and Catherine for their friendship over the past two years.

And last, but definitely not least, I would like to thank my precious family and Jeff who is truly my best friend. This acknowledgement will not do justice to how grateful I am for your love and support. Thank you for your prayers and encouragement when I needed it most. I would not be where I am today without you.

## Table of Contents

Approval Page.....	ii
Acknowledgements.....	iii
Table of Contents.....	iv
List of Figures.....	viii
List of Tables.....	x
List of Abbreviations.....	xii
Abstract.....	xv
Chapter 1 Introduction.....	1
Chapter 2 Review of Literature.....	7
2.1. Dietary Antioxidants.....	7
2.1.1. Beneficial Effects of Consumption of Dietary Antioxidants....	8
2.1.2. Sources of Dietary Antioxidants.....	10
2.2. Estimation of Usual Intakes among Populations.....	11
2.2.1. Misreporting of Nutrient Intake.....	12
2.2.2. Within- and Between-Person Variation of Nutrient Intake...	15
2.2.3. Average Macronutrient Intake.....	18
2.2.4. Average Vitamin and Mineral Intake.....	19
2.2.5. Average Antioxidant Intakes.....	21
2.3. The Number of Days Required to Accurately Assess Nutrient Intake.....	22
2.4. Dietary Reference Intakes for Assessing Adequate Nutrient	

Intake.....	25
2.4.1. Adequacy of Daily Nutrient Intake among Young Adults: Are They Meeting the Guidelines?.....	28
2.4.2. Lifestyle Factors Associated with Nutrient Intake among College Students.....	31
2.5. Supplement Use among U.S. Population Groups.....	32
2.5.1. Supplement Type and Frequency.....	34
2.5.2. Contribution of Supplement to Nutrient Adequacy.....	35
Chapter 3 Materials and Methods.....	41
3.1. Study Design and Population.....	41
3.2. Dietary Assessment.....	42
3.3. Nutrient Analysis.....	43
3.4. Misreporting Nutrient Intake.....	45
3.5. Statistical Analysis.....	47
Chapter 4 Results.....	52
4.1. Misreporting and Usual Intake for Energy and Key Nutrients among College Students.....	52
4.1.1. Under- and Over-reporting.....	52
4.1.2. Lifestyle Characteristics of Participants who were Average Reporters.....	53
4.1.3. Mean Intakes and Variance Components.....	54
4.2. The Number of Days to Accurately Assess Usual Intakes for Energy And Key Nutrients, Including Antioxidants, among College Students.....	57

4.2.1. Effects of the Number of Dietary Recalls on Intake Distributions.....	57
4.2.2. Number of Days of Dietary Assessment Required to Assess Usual Antioxidant Intake.....	58
4.3. Usual Nutrient Intake from Diet and from Supplement Adequacy Assessed by the EAR Cut-Point Method among College Students.....	59
4.3.1. Nutrient Intake Adequacy among Genders.....	59
4.3.2. Supplement Types and Use.....	60
Chapter 5 Discussion.....	79
5.1. Identification of Misreporting and Estimation of Usual Intakes for Energy and Key Nutrients, Including Antioxidants, among College Students.....	79
5.1.1. Misreporting among Male and Female College Students....	79
5.1.2. Mean Intakes of Energy and Key Nutrients among Male and Female College Students.....	81
5.1.3. Within- and Between-Person Variation in Nutrient Intakes of Male and Female College Students.....	84
5.2. The Number of Days to Accurately Assess Usual Intakes for Energy And Key Nutrients, Including Antioxidants, among College Students.....	89
5.3. Assessment of Nutrient Intake Adequacy and Impact of Supplement Use on Nutritional Status of College Students.....	90
5.3.1. Lifestyle Factors among College Students.....	91
5.3.2. Nutrient Intake Adequacy According to the EAR Cut-Point	

Method.....	92
5.3.3. Supplement Usage and Contribution to Intake Adequacy among College Students.....	96
5.4. Conclusion and Future Direction.....	99



## List of Figures

<b>Figure 4.1</b> Daily intakes for three college students from UCONN at the 10 <sup>th</sup> , 50 <sup>th</sup> , and 90 <sup>th</sup> percentiles of distribution for total fat intake (A) and total flavonoid intakes (B).....	65
<b>Figure 4.2-1.</b> Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of energy intake for 44 college students from UCONN. Mean energy intake ( $\pm$ S.D.) for 1 day was 2,216 $\pm$ 720 kcal/d, 2,280 $\pm$ 642 kcal/d for 7 days, and 2,274 $\pm$ 583 kcal/d for 30 days, respectively.....	70
<b>Figure 4.2-2.</b> Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of vitamin C intake for 44 college students from UCONN. Mean vitamin C intake ( $\pm$ S.D.) for 1 day was 107.3 $\pm$ 76.8 mg/d, 117.6 $\pm$ 64.9 mg/d for 7 days, and 130.9 $\pm$ 52.7 mg/d for 30 days, respectively.....	71
<b>Figure 4.2-3.</b> Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of total flavonoid intake for 44 college students from UCONN. Mean total flavonoid intake ( $\pm$ S.D.) for 1 day was 236.4 $\pm$ 449.9 mg/d, 177.2 $\pm$ 282.5 mg/d for 7 days, and 165.7 $\pm$ 201.7 mg/d for 30 days, respectively.....	72

**Figure 4.2-4.** Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of isoflavone intake for 44 college students from UCONN. Mean isoflavone intake ( $\pm$ S.D.) for 1 day was  $3.9\pm12.8$  mg/d,  $5.1\pm7.8$  mg/d for 7 days, and  $4.5\pm5.3$  mg/d for 30 days, respectively.....73

**Figure 4.3.** The number of days of dietary assessment required to estimate usual macronutrient and antioxidant intakes among total, male, and female college students from UCONN after adjusting for energy intake and gender for total population and energy intake among genders,respectively.....74

**Figure 4.4.** Form of supplement consumed by 17 male and female supplement users from UCONN.....77

## **List of Tables**

<b>Table 2.1.</b> A comparison of micronutrient intakes by gender and age.....	39
<b>Table 4.1.</b> Characteristics and misreporting of 60 male and female college students from UCONN.....	63
<b>Table 4.2.</b> Lifestyle characteristics of 44 male and female college students from UCONN.....	64
<b>Table 4.3.</b> Means, coefficients of variation, and variance ratios for energy and nutrient intakes among 44 college students from UCONN.....	66
<b>Table 4.4.</b> Means, coefficients of variation, and variance ratios for energy and nutrient intakes among 44 male and female college students from UCONN.....	67
<b>Table 4.5.</b> The number of days of dietary assessment required to estimate usual micronutrient intakes among male and female college students from UCONN..	75
<b>Table 4.6.</b> Usual nutrient intake adequacy for select nutrients among 44 male and female college students from UCONN.....	76

<b>Table 4.7.</b> Comparison of usual nutrient intake adequacy between dietary supplement users and non users from UCONN.....	77
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### **List of Abbreviations**

ACS	American Cancer Society
ACSM	American College of Sports Medicine
ADA	American Dietetic Association
AHA	American Heart Association
AI	Adequate Intake
AMDR	Acceptable Macronutrient Distribution Range
BMI	body mass index
BMR	basal metabolic rate
CAD	coronary artery disease
CHD	coronary heart disease
CSFII	Continuing Survey of Food Intake of Individuals
CVD	cardiovascular disease
DFE	dietary folate equivalent
DGA	Dietary Guidelines for Americans
DR	dietary recalls
DRI	Dietary Reference Intakes
DSHEA	Dietary Supplement Health and Education Act
EAR	Estimated Average Requirement
EI	energy intake
EPIC	European Prospective Investigation in Cancer and Nutrition
FAO	Food and Agriculture Organization of the United Nations

FCS	Food Consumption Survey
FLDB	USDA flavonoid database
FFQ	Food Frequency Questionnaire
INTERMAP	International Study of Macro- and Micro-nutrients And Blood Pressure
IOM	Institute of Medicine
NAS	National Academy of Sciences
NCC	Nutrition Coordinating Center
NCHS	National Center for Health Statistics
NDSR	Nutrition Data System for Research
NGHS	The National Heart, Lung, and Blood Institute Growth and Health Study
NHANES	National Health and Examination Survey
NHEFS	National Health and Nutrition Examination Survey Epidemiologic Follow Up Study
PAL	physical activity level
RAE	retinol activity equivalents
RDA	recommended dietary allowance
RNS	reactive nitrogen species
ROS	reactive oxygen species
SAS	statistical analysis systems
SD	standard deviation
SU.VI.MAX	Supplémentation en Vitamines et Minéraux Antioxydants

TE	tocopherol equivalents
UConn	University of Connecticut
UL	Upper Limit
USDA	United States Department of Agriculture
WHO	World Health Organization

## **Abstract**

High intake of antioxidant rich foods has been shown to decrease risk factors of chronic disease. Young adulthood may be crucial in establishing healthy lifestyles including adequate nutrient consumption.

The present study was designed 1) to estimate usual nutrient intakes, 2) to calculate the number of days required to estimate usual antioxidant intake, and 3) to assess intake adequacy from diet and diet + supplement sources by using the Estimated Average Requirement (EAR). The USDA Flavonoid and Proanthocyanidin databases, food consumption data, and dietary supplement use data from 60 students aged 18-25 years at the University of Connecticut were utilized.

After applying the Goldberg cut-off equation defined for this population, 27% of participants were classified as misreporters of intake. Males consumed higher mean intakes than females for 13 of the 27 nutrients after adjusting for energy intake ( $P < 0.05$ ). After adjusting for energy and gender, a 7-day dietary recall was adequate to achieve  $r \geq 0.9$  for fat, carbohydrate, protein, lycopene, and proanthocyanidin. More than 40% of females had intakes below the EAR for vitamins D and E, calcium, and magnesium. With the addition of a supplement, supplement users consumed more for all nutrient intakes except vitamin A ( $P < 0.05$ ). Nutritional adequacy of users improved for vitamins D and E, and magnesium compared to non-users ( $P < 0.05$ ). Overall, more than 7 days would be required to estimate usual nutrient intakes, students were consuming intakes



below adequacy for most nutrients, and supplement usage increased nutrient intake and adequacy compared to nonusers.

## **Chapter 1**

### **Introduction**

Dietary behaviors and patterns of physical activity established in young adulthood are at the foundation for development or prevention of diseases that often do not manifest until late adulthood (1-3). Lifestyle factors that affect the risk of obesity and chronic diseases may begin in early adolescence and continue to increase during this transition into adulthood (4, 5). This period of development can be crucial in establishing healthy lifestyles that include adequate consumption of vital nutrients and physical activity (6, 7). Increased consumption of fruits and vegetables has been shown to decrease risk factors associated with the development of chronic diseases (8, 9) such as cardiovascular disease (CVD) (10, 11), certain cancers (12, 13), type 2 diabetes (14), and other degenerative diseases (15, 16). Several studies provide evidence to support that the high concentration of antioxidants, such as vitamin C, vitamin E, carotenoids, and flavonoids, in these foods may be responsible for reducing risk factors (17-19). The essentiality of many macro- and micronutrients in preventing deficiencies requires a dietary recommendation for intake at a specified level for adequacy in the general population. This is incorporated in the dietary reference intakes (DRIs) by the Institute of Medicine (IOM) (20). However, flavonoids and other polyphenolic compounds are still considered under review mainly due to the lack of data on comprehensive food composition and bioavailability (21).

Young adults today have had the benefit of growing up under the influence of dietary guidelines and national goals to improve health (22, 23). The campus of a university or college can provide an environment of intellectual growth as well as healthful dietary behaviors. Many campuses provide access to gyms, workout classes, dietary counseling, healthy options at the dining halls, nutrition classes, and more. A previous study suggested that college students compared to non-student counterparts overall had lower risk factors associated with chronic diseases due to lifestyle choices (24). According to a recent review, 96% of U.S. young adults are considered to be in good health measured by traditional standards (2). However, in the same review, it was reported that young adults aged 18-24 y have twice the mortality as adolescents aged 12-17 y. In addition, other studies have shown that many students are not meeting many of the established dietary guidelines or recommendations for adequate nutrient intakes and physical activity (25-27). Data show that place of residence, new academic and social pressures, weight concerns, skipping meals, and access to fast food are a few contributing factors to inadequate nutrient intakes (24, 28-30). Failure to meet daily requirements for fruit, vegetables, dairy products, whole grains, and physical activity can put individuals at greater risk for nutrient inadequacy or deficiencies which may increase risk factors associated with chronic diseases (31, 32). The use of dietary supplements in the U.S. is becoming more prevalent and is continuing to increase specifically among adults aged 20 y and older (33, 34) and may play an important role in nutrient intake adequacy (35). However, the American Dietetic Association (ADA) recommends that only individuals who

restrict energy intake or are on a severe weight loss diet, eliminate a food group from usual diet, or who consume high carbohydrate and low micronutrient dense diets should use supplementation (36).

Estimation of usual intake of a population is essential in the process of establishing the protective effects of nutrients against the development of certain diseases and assessing nutrient intake adequacy (37). Common assessment tools used in epidemiological studies include diet histories, dietary recalls (DR), food frequency questionnaires, and diet records (38). A long term assessment of daily intakes is required to assess usual intakes; however, due to the cost and burden of this requirement, most studies employ shorter term assessments. Many limitations exist with dietary assessment tools which can alter the results and conclusions of intake data (39). Examples of important limitations include misreporting nutrient consumption (40) and large within- and between-variation in daily intakes of participants (41) which can weaken the relationship between dietary intake and disease risk factors. Underreporting of nutrient intake is a significant problem in nutritional epidemiological studies and may increase the estimation of inadequate nutrient consumption as well as affect the interpretation of nutrient distributions and its applications to a population group (42). Daily intakes can vary greatly from day-to-day often as a result of seasons or cultural or environmental factors (43). This within-person variation, as well as the variation between individuals, must be estimated to understand the relationship of diet and health status. Statistical methods have been developed to control for these factors to produce accurate estimates of usual intake from shorter

recording periods (37, 44-46); however, increasing the number of days of dietary assessment greatly decreases the bias associated with this source of variation (41). In addition, the number of days of diet record necessary to accurately estimate true intake for each nutrient should be carefully considered when designing a study for a specific population group (47, 48).

In comparison to adolescents and adults, limited data is available on the nutrient adequacy and supplemental intake in young adults that includes assessment of antioxidant intakes. In addition, there are few studies that evaluate research methodology for assessing variation and intake of nutrients and non-nutrient antioxidants in the U.S. diet. Therefore this study was conducted to assess nutrient adequacy from diet and supplement and to estimate usual antioxidant intakes in well-educated, healthy young adults.

**Objective 1: Identify misreporting and characterize the variation of nutrient intakes among U.S. college students**

The goal was 1) to validate the dietary assessment data collected over 30 consecutive days by identifying misreporting of energy intake among a subset population of college students, and 2) to describe the mean, the within- and between-person variation, and the variance ratios of nutrient intake among healthy college students. The working hypotheses included: (H<sub>1</sub>) more students would be identified as underreporters than overreporters; (H<sub>2</sub>) more females would underreport energy intake than males; and (H<sub>3</sub>) the micronutrient, including antioxidants, intakes have greater day-to-day variability than

macronutrients resulting in larger within-person variation and higher variance ratios.

**Objective 2: Determine number of days required for assessing usual antioxidant intakes in diet of U.S. college students**

The goal was to utilize the within-and between-variation values of macronutrient and antioxidant intakes to calculate the number of days of DR would be required to assess the truest intake among the same subset college population. In addition, the effects of sampling shorter recording periods common to many nutritional epidemiological studies on the distribution of intakes were compared. Therefore, the working hypotheses were: ( $H_1$ ) increasing the number of days of dietary assessment decreases the variation; and ( $H_2$ ) antioxidants require more days of dietary assessment than macronutrients to estimate usual intake in this population.

**Objective 3: Assess nutrient adequacy from diet and supplement sources among U.S. college students**

The goal was 1) to assess intake adequacy by determining male and female students whose usual nutrient intakes fell below the most recent Estimated Average Requirement (EAR) (49) using the EAR cut-point method suggested by the Institute of Medicine (IOM) (20), and 2) to evaluate supplement use and contribution to overall nutrient adequacy among users. The working hypotheses included: ( $H_1$ ) females are consuming lower intakes of nutrients and therefore, their diet is more inadequate than males; ( $H_2$ ) more females than males consume supplements consistently with a multivitamin supplement being the most

prevalent; and (H<sub>3</sub>) supplementation significantly increases nutrient adequacy among users compared to non-users.

## **Chapter 2**

### **Review of the Literature**

#### **2.1. Dietary Antioxidants**

Chronic diseases remain among the major causes of death in the United States. The most recent statistics from the American Heart Association (AHA) and the American Cancer Society (ACS) report that over 81 million Americans have one or more types of cardiovascular disease and that 569,490 deaths from cancer were projected in 2010 (50, 51). Increased consumption of fruits and vegetables has been shown to decrease risk factors associated with the development of chronic diseases (8, 9) such as cardiovascular disease (CVD) (10, 11), certain cancers (12, 13), type 2 diabetes (14), and other degenerative diseases of aging such as cognitive diseases and decreased immune function (15, 16). In the U.S., National Health and Nutrition Examination Survey (NHANES) has been conducted to obtain health and nutritional information from interviews, bio specimens, 24-h DR, and questionnaires from over 8000 individuals (52, 53). The National Health and Nutrition Examination Survey Epidemiologic Follow Up Study (NHEFS), is an ongoing prospective cohort study that includes participants ages 25-74 y from the first NHANES collected in 1971-1975. Bazzano et.al. reported results from the NHEFS among the participants with no history of CVD at baseline and the relationship between fruit and vegetable intake and CVD incidence (54). Overall, participants consuming three or more servings of fruits and vegetables as compared to less than one serving was associated with a 27% lower stroke mortality, a 42% lower ischemic heart



disease mortality, a 24% lower ischemic heart disease mortality, a 27% lower CVD mortality, and a 15% lower all cause mortality. The Women's Health Study, with 34,000 postmenopausal U.S. women, (11) reported a dose dependent inverse relationship with fruit and vegetables and the relative risk of CVD while the Physicians' Health Study in men (55) reported a decrease in coronary heart disease (CHD) with a higher consumption of vegetables rich in carotenoids. In relation to cancers, a meta-analysis of epidemiologic studies identified that the case-controlled studies included supported a significant decrease in risk factors associated with esophageal, lung, stomach, and colorectal cancers with an increase consumption of fruits and vegetables (56). Several studies provide evidence to support the hypotheses that the high concentration of antioxidants, such as vitamin C, vitamin E, carotenoids, and polyphenols, in fruits and vegetables may be responsible for reducing risk factors (17-19).

#### *2.1.1. Beneficial Effects of Consumption of Dietary Antioxidants*

These dietary antioxidants function in human metabolism to reduce or prevent the production of reactive oxygen species (ROS) or reactive nitrogen species (RNS) that generate oxidative stress by producing free radicals. An accumulation or overexposure to these reactive species can damage DNA, proteins, and lipid membranes which has been linked to the development of many chronic diseases (57). Vitamin E is a fat soluble vitamin, therefore, it protects lipid from peroxidation by functioning as a chain breaking antioxidant. The term vitamin E actually represents 8 different compounds, four tocopherols and four tocotrienols (58). Alpha-tocopherol is the only vitamer among the

tocopherols with an established dietary recommendation (59). Vitamin C functions as a reducing agent by becoming oxidized to prevent free radical damage from ROS or RNS. Carotenoids, specifically  $\beta$ -carotene, can react with singlet oxygen species to prevent oxidative damage (58). While supplementation trials of these nutrients have reported inconsistent results regarding the reduction of disease risks (60-63), studies analyzing consumption from dietary sources yield more promising results (64-67).

Flavonoids are the most common and largest plant polyphenolics present in plant sourced diets with over 6000 different flavonoids identified. Flavonoids, all with a 3 carbon ring structure, exist in 6 major classes: flavanones, flavones, flavonols, flavanols, anthocyanins, and isoflavones (68). Flavonoids are hypothesized to be radical scavengers by interacting with highly reactive free radicals creating a stable flavonoid radical. Therefore, consumption of foods rich in flavonoids, as well as other polyphenols, may prevent endogenous antioxidants from being oxidized. These hypotheses give flavonoids antioxidant properties that may protect vascular and cardiovascular function (69). A review by Khan et.al in 2008 concluded that various polyphenols such as resveratrol, the isoflavone genistein, and certain flavanols, as well as the carotenoid lycopene, modulate many of the signal transduction pathways in the metastasis of cancers that include skin, breast, prostate, lung, and liver in vitro (18). While clinical trials are needed to determine effects in prevention and treatment of cancers in humans, this evidence is promising for future research. According to a review of epidemiological studies including data on polyphenols and the relationship with

disease, 7 of the 12 cohort studies found protective effects of flavones, flavonols, and/or catechins with relation to coronary artery disease (CAD) (70). A few of these studies included: the Rotterdam study that found an inverse association between tea intake and myocardial infarction among 4800 men and women (71); the Zutphen Elderly Study that concluded both intake of catechins from tea could explain the reduction in ischemic heart disease in men after a 10 y follow up and that dietary flavonols decreased risk for CHD (72-74); and a study who utilized the participants from the Alpha-Tocopherol, Beta-Carotene Cancer Prevention Study that reported an inverse association with flavonols and flavones and risk of CVD among male smokers (75). In conclusion, dietary antioxidants have been shown to have beneficial effects on the risk factors for chronic diseases.

#### *2.1.2. Sources of Dietary Antioxidants*

Dietary antioxidants are ubiquitous in nature and major sources are the deeply pigmented fruits and vegetables. Examples of vitamin C rich foods include citrus fruits, strawberries, bell peppers, and broccoli (76). Nuts, seeds, and oils are rich sources of vitamin E due to its fat soluble properties (77). Carotenoids include over 600 total compounds, however, only six are important in human metabolism:  $\alpha$ -carotene,  $\beta$ -carotene,  $\beta$ -cryptoxanthin, lycopene, lutein, and zeaxanthin. Major sources of these compounds include yellow and green leafy vegetables, carrots, red peppers, oranges, tomatoes, and egg yolk (78). Flavonoids provide yellow pigments in bell peppers, celery, and onions as well as the red color in grapes and plums. The subclass flavanols are commonly found in green tea, apples, and red wine. Isoflavones exist mainly in soy beans (79).

Proanthocyanidins, which are polymers of flavan-3-ols, can be found in berries, chocolate, beans, and cinnamon (80). Major sources of these polyphenolic compounds in the U.S. diet are teas, citrus fruits and juices, and wine as well as green leafy vegetables and fruits such as apples and berries (81, 82). Dietary antioxidants are highly concentrated in specific foods or food groups. Therefore, estimation of intakes in a population is the next vital step in establishing their protective effects against chronic disease (82).

## **2.2. Estimation of Usual Nutrient Intake among Populations**

Estimation of usual or habitual nutrient intake of a population is a vital process in nutritional research. The truest representation of intake for an individual collectively defines usual intake for a population (39). These values are required in order to: define nutrient intake adequacy, contribute to the baseline data required for dietary guidelines for specific age groups, provide a basis for nutritional interventions, and establish the relationship between diet and health status concerning malnutrition or risk of disease (38). Considerable effort has been devoted to analyzing the effects of this latter point, specifically in relation to cancers and CVD as stated in earlier sections. Unfortunately, only a limited few have dedicated research to defining the most accurate method to determine these habitual dietary intakes (41, 45, 46, 83, 84). Common assessment tools used in epidemiological studies include diet histories, DR, food frequency questionnaires (FFQ), and diet records (85). Long term dietary assessments are required to provide an accurate representation of dietary

intakes and patterns. DR or diet records are often employed to assess usual intake, however, their use of weeks or even months is rarely employed due to participant responsibility and errors associated (39). Therefore, many studies report nutrient intakes using shorter record periods which may provide less reliable data (86). Despite the advances in technology to collect nutrient data from these assessment tools, limitations exist. Major limitations include misreporting of nutrient consumption and variation of intakes from day-to-day and between-individuals which all create inconsistent results and gaps in the relationship between diet and disease (87, 88).

#### *2.2.1. Misreporting of Nutrient Intake*

Misreporting by all population groups is a serious issue in nutrition and health related research. It can compromise the accuracy, validity, and application of data reporting any nutrient intake. While collection of long term dietary intakes is preferred, it can also increase the likelihood of misreporting intake due to the burden of daily recording accurate intakes and the first day of any diet recording period is considered to be the most accurate (45, 89). The term misreporting encompasses under-reporting and over-reporting nutrient intake (40). Underreporting or low energy reporting can be the result of under estimating food intake, elimination of certain foods or amounts, or under-eating due to dietary restrictions or dieting (90). Regardless of classification of misreporting, the inclusion of individuals who provide inaccurate nutrient intakes can alter the results and conclusions significantly. A common method for identifying such individuals is the Goldberg's cut-off equation (91). This equation

requires the average energy intake, average basal metabolic rate, and daily physical activity of the population to generate critical values for energy intake applied to the average energy intake of each individual participant. The population critical values are represented by energy intake: BMR estimated ratio ( $EI_{rep}:BMR$ ).

A review by Black provides a guide for the use of the Goldberg cut off in nutritional assessment research (40). In the review, Black emphasizes the importance of selecting a physical activity level (PAL) for each population dependent on reported daily physical activity and classifications provided by the World Health Organization (WHO) (92). For the average student population group, a 1.6-1.7 PAL is suggested for determining energy requirements (92). In data reported by Black (40), a PAL of 1.7 was used for young adults aged 18-29 y who were predominately non Hispanic White and participated in moderate leisure activities. However, a high PAL value can inaccurately identify individuals as low energy reporters especially in a study with a small sample size (40). In order to increase the sensitivity and specificity of the Goldberg cut off in a study with a population size ( $n$ ) < 100, the number of days of dietary intake assessment should be increased (93). However with larger sample sizes, a fewer number of dietary assessments may be used. Results from NHANES III for misreporting using the Goldberg cut off include a critical value of 0.9 to 1.54 with a mean  $EI_{rep}:BMR$  of 1.36 for all adults. In addition, 18% of males and 28% of females were classified as underreporters of energy intake (42).

After the identification of misreporters in a population group, causality should be determined before the decision to include or exclude individuals from the results. In a recent review, Poslunsna et. al. summarized the main causes of errors in 24-hr DR and food records most frequently reported in 38 nutritional studies (94). Results indicated that the major determinants for misreporting included: BMI, age and sex, socioeconomic status and education, health related activities, psychological factors, and eating habits. While misreporting includes both underreporting and overreporting of nutrient intake, overreporting was identified less frequently in these studies. The most consistent factor reported in the review was that as BMI increased, a larger percentage of the population was classified as misreporters. In addition, more females than males tended to misreport their nutrient intake (94). Similar gender results were found in a study with 53 non obese, weight stable adults. They reported 49% of the females and 14% of the males were identified as underreporters from a 7-day DR (89).

It remains unknown whether males tend to underreport less than women or if their higher energy requirements allow them to rarely fall below the cutoff limits when applied to an entire study population (93); however, Asbeck et. al. reported that the higher percentage of female underreporters was due to restrained eating practices evidenced by scores from an eating practice survey in a normal weight population (89). Leibman et. al. conducted a study with 324 college students analyzing the relationship between dieting practices, gender, and psychological variables such as self image and body perceptions (95). Results reported were that 38% of females and 13% of males has dieted to lose

weight within the past year and more females reported patterns of disordered eating, such as fat avoidance or replacement, and body dissatisfaction (95). Body weight dissatisfaction, frequent dieting, and societal pressures seem to be an area of concern in young adult and adult female populations; therefore the validity of dietary assessments from these population groups should be analyzed before average intake results are reported (96).

#### *2.2.2. Within- and Between-Person Variation of Nutrient Intake*

Day-to-day variability in nutrient intakes can significantly alter the statistical outcomes and interpretations of dietary assessment data. This fluctuation is defined as within-person variation and can be attributed to environmental and cultural factors (97). Micronutrients have a higher concentration in specific foods and tend to have greater variation due to seasonal variation or the wide array of food choices available in many developed countries when compared to macronutrients which remain more stable in the diet. However, seasonal variation has a greater impact in developing countries where all foods are not as easily accessible (97). Day of the week sampled by a dietary assessment tool is another source of within-person variation. Energy and protein consumption are typically larger on the weekends compared to the weekdays and should be considered when using 24-hr DR (45). Within-person variation can be estimated and must be adjusted for statistically due to its high correlation to the mean of the sample day. This is crucial in the interpretation when the study design only includes a small number of days of dietary intake (41). However, increasing the number of days of diet recorded can decrease the



within-person variation significantly (39). Another important consideration is a large variation between individuals of a population because it may misconstrue the relationship between nutrient consumption and disease risks (40, 98). Between-person variation can be reduced by accounting for certain demographics and lifestyle factors specific to the group of study (48, 97, 99). The ratio of the within-to-between variation can be used to further describe the effect of the within-person variation as the greater the variance ratio, the greater the within-person variation in daily intakes (37).

Several methods have been developed to assess usual dietary intake among populations (37, 44-46); however all methods require estimation of within- and between-person variation. Therefore, these values must be calculated from multiple number of diet records or values can be borrowed from an appropriate subset population (37). Chang et. al. analyzed the within- and between-person variation among Taiwan college students who completed a total of three 5-day DR (37). They found that males had larger within to between ratios for fat, protein, polyunsaturated fatty acid, vitamin A, thiamin, and riboflavin than the females which they attributed to the irregular eating patterns and possible bingeing of male college students. Females had larger within-person variation in the intakes of carbohydrates which could be a result from the common practice of dieting or meal skipping in this population group (37). In another study, Jahns et. al. analyzed the effects of gender as well as age and culture on the estimation of within- and between-person variation in U.S. and Russian older children and adolescents (99). Results were reported from nonconsecutive 24-hr DR from the

Russia Longitudinal Monitoring Survey (RLMS) and the Continuing Survey of Food Intake by Individuals (CSFII). They analyzed energy intake and 10 additional macro- and micro-nutrients: protein, carbohydrate, fat, calcium, iron, magnesium, thiamin, riboflavin, niacin and the antioxidant nutrient as vitamin C. Among the U.S. population, they found that the girls had higher within-person variation than the boys for all nutrients excluding carbohydrates and the girls had higher between-person variation as well. Results pertaining to the differences in age groups reported that the older Russian girls had higher within-person variation for all nutrients except riboflavin, niacin, and vitamin C as well as higher between-person variation for all nutrients except magnesium and thiamin than the younger girls. No observable patterns was found among the U.S. age groups for within-person variation but the between-person variation was higher for the older girls for 9 out of the 11 nutrients including vitamin C (99).

In U.S. men and women, Neuhaus et. al. analyzed the ratios of within-person variation to between-person variation in different age groups for energy, 3 macronutrients, and 9 micronutrients including vitamin C (100). They found that as age increased, the variance ratio decreased meaning the within-person variation approached the between-person variation. These results were significant among the men for most nutrients, however, a decreasing trend was not as apparent for the women (100). Overall, the results seem to indicate that younger adult populations may have larger day-to-day variability in nutrient intake which has important implications with estimating usual nutrient intakes of a population. While these studies do include within- and between-person variation

among adolescents and young adults, there is a gap in the literature pertaining to antioxidant intakes among this age group in the U.S.

### *2.2.3. Average Macronutrient Intake*

In a report, *What We Eat in America*, the United States Department of Agriculture (USDA) summarizes nutrient intake according to gender and age from analyses of the NHANES (101). Results from NHANES 2007-2008 for energy and macronutrient intakes among males aged 12-19 y and 20-29 y include on average: 2,424 kcal and 2,756 kcal energy, 90.7 g and 105.3 g protein, 313 g and 342 g carbohydrate, and 90.6 g and 96.4 g fat, respectively. In the same report, results for the females aged 12-19 y and 20-29 y were as follows: 1861 kcal and 1828 kcal energy, 65.6 g and 68.3 g protein, 248 g and 231 g carbohydrate, and 69.2 g and 67.5 g fat, respectively (101).

Average nutrient intakes from the students at the University of New Hampshire (27) included: average caloric intake for the males and females was  $2,740 \pm 842$  and  $1,879 \pm 547$  kcal/d, respectively and carbohydrate intakes of  $343 \pm 113$  g/d for the males and  $254 \pm 78$  g/d for the females. Almost 100% of the population was within the guideline for protein intake with intakes of  $118 \pm 47$  g/d and  $73 \pm 24$  g/d for males and females respectively; however fat intake varied depending on gender. Average fat intake for the males was  $93 \pm 35$  g/d while the average fat intake for the females was  $63 \pm 26$  g/d (27).

U.S. dietary intakes were similar when compared to other countries. In 2004, Lambert et. al. collected and evaluated data on the nutrient intake of European children and adolescents (102). They included 79 surveys from 23

countries in the review that reported intake data for energy, protein, fat, carbohydrates, vitamins, minerals, and trace elements. Results revealed that energy intake increased with age but reached a level intake with older adolescents. Older adolescent males were reported as consuming an average intake of 9,000-16,500 kJ/d (2,151-3,943.5 kcal) for energy and older adolescent females consumed from 6,800 to 10,600 kJ/d (1,625-2,533 kcal). Protein intake for males and females increased with age with the older adolescents reporting the highest average intake. Males 15-18 y of age reported 71-127 g/d while females reported 53-88 g/d (102).

#### *2.2.4. Average Vitamin and Mineral Intake*

Table 1 shows a comparison of micronutrient intakes from adolescents and young adult populations in the U.S., Europe, and Japan (27, 101-103). Among the NHANES 2007-2008 participants, males in general had higher intakes than the females except  $\alpha$ -carotene,  $\beta$ -carotene,  $\beta$ -cryptoxanthin, and lutein + zeaxanthin among the 20-29 y age group (101). In the same study, the older males consumed slightly higher average amounts of micronutrients than the younger males except for vitamin A,  $\alpha$ -carotene, lutein + zeaxanthin, vitamin D, and calcium; however, all average nutrient intakes among the males were similar. The same intake trend was reported among the older females as well (excluding thiamin, folate, vitamin D, calcium, and iron) (101). Nutrient intake results continued from the University of New Hampshire college students, reported by Burke et.al. (27) were found to be higher than the NHANES 2007-2008 participants for both genders for vitamin A, vitamin C, calcium, and

potassium. However, the standard deviations reported by Burke et.al. were quite large. The female participants from NHANES 2007-2008 consumed a higher amount of folate than the U.S. college female population. In general, the U.S. populations were within similar intake range for most nutrients.

In the same European review as previously mentioned (102), Lambert et.al. reported micronutrient intakes in average ranges among children and adolescents separated by age and gender from various countries in Europe. In general, U.S. average intakes were within the ranges according to gender. For NHANES 2007-2008 male participants, average intakes of thiamin, riboflavin, and vitamin B<sub>6</sub> were higher than the European adolescents. However, the male and females had lower average intakes than the European adolescents for magnesium and potassium. The females from the NHANES reported lower intakes for niacin only. Burke et.al. reported higher average intakes for both male and females for vitamin C when compared to the European population (27). In comparison to the Japanese college females vitamin intakes reported by Kimura et.al. (103), the females from the NHANES reported similar intakes of niacin, vitamin B<sub>12</sub>, vitamin C, and vitamin E. However, the Japanese females consumed higher intakes for vitamin A and vitamin D. The remaining nutrients were consumed in lower amounts than the U.S. females. In comparison to the U.S. college females, the Japanese females consumed lower average intakes for vitamin A, vitamin C, and vitamin D but still within a similar range. Overall, vitamin and mineral intakes were similar across countries and male average

intake was higher than females for this young adult population group (27, 101-103).

#### *2.2.5. Average Antioxidant Intakes*

Taking a closer look at specific antioxidant nutrients, Chun et.al. reported antioxidant intakes from diet and supplement from NHANES 1999-2002 intake data (104). Adults aged 19-30 y reported intakes of  $96.5 \pm 4.2$  mg/d vitamin C,  $6.8 \pm 0.1$  mg/d vitamin E,  $143.5 \pm 9.3$   $\mu$ g/d *RAE* carotenes, and  $189.9 \pm 18.0$  flavonoids. Intakes varied according to gender with males consuming more daily than females except for carotenes. Males consumed average intakes of  $104.6 \pm 3.4$  mg/d vitamin C,  $8.0 \pm 1.0$  mg/d vitamin E,  $185.9 \pm 8.1$   $\mu$ g/d *RAE* carotenes, and  $214.1 \pm 13.8$  mg/d flavonoids. Females consumed average intakes of  $86.6 \pm 2.7$  mg/d vitamin C,  $6.2 \pm 0.1$  mg/d vitamin E,  $198.6$   $\mu$ g/d carotenes *RAE*, and  $200.2 \pm 12.1$  mg/d flavonoids (104). From the same survey, isoflavones were consumed by only 35% of the adult population who reported a mean intake of 3.1 mg/d which results in a 1.0 mg/d mean intake for all adults (105). Proanthocyanidin intake among adults 19-30 y was  $81.4 \pm 6.8$  mg/d with a mean intake of 95 mg/d for the total population (80).

In comparison, the antioxidant intakes among a Greek subset population, who participated in the European Prospective Investigation in Cancer and Nutrition (EPIC), were higher than the U.S. adults apart from polyphenol intake (106). Average intakes for the total population were as follows: 214 mg/d vitamin C, 28 mg/d vitamin E, 4,660  $\mu$ g/d  $\beta$ -carotene, 92 mg/d flavonoids, <0.1 mg/d isoflavone, and 75 mg/d proanthocyanidin. Males had higher intakes for all

antioxidants except isoflavones which was only presented as  $<0.1$  mg/d for both genders. Intakes for males and females were 220 and 209 mg/d vitamin C, 31 and 26 mg/d vitamin E, 4,828 and 4,532  $\mu\text{g/d}$   $\beta$ -carotene, and 89 and 67 mg/d proanthocyanidins, respectively (106). The National Nutrition Survey in Australia reported age related flavonoid intake from 17,326 individuals. Average flavonoid intake from adults  $> 18$  y was 454 mg/d (107). A Danish Household Consumption Survey reported 175 mg/d of total flavonoids which was similar to the U.S. flavonoid data (108), while the Dutch National Food Consumption Survey reported higher intakes at 211 mg/d (109). Overall, antioxidant consumption is dependent on dietary habits and behaviors of an individual and of a country. The average intake of antioxidants varies depending on the age, gender, origin of the source and other lifestyle characteristics (80, 82, 104); however, a similar trend of intakes is evident.

### **2.3. The Number of Days Required to Accurately Assess Nutrient Intake**

It is important, when developing a study design, to know how many days of dietary assessment is required to produce accurate and reliable intake results for a population group (47). To assess usual nutrient intake levels among a population, within- and between-person variation should be estimated and included in a calculation to determine sufficient number of diet record necessary to produce accurate results (47, 48).

The calculation of days (D) of nutrient intake includes the ratio of within-person variation ( $S_w$ ) to between-person variation ( $S_b$ ) (110). The variability in

daily nutrient intake among adults has been shown to be greater than the variability between individuals in a study population (47), and the smaller the ratio, the fewer number of days is required to estimate the nutrient intake within a specified level of accuracy ( $r$ ) between the true intakes and the observed intakes (47). Nelson et. al. analyzed data from 18 studies that reported mean nutrient intake, values for within- and between-person variation, and the number of days required to estimate true intake within a give accuracy. They included studies with populations aging from infancy to older adults and reported a total of 29 nutrients including energy. Values presented for D were based on  $r = 0.9$ . Most nutrients required more than 7 days of DR to estimate true intake in all age and gender groups. Among the adult populations, energy, protein, carbohydrate, and fat required 4-8 days depending on gender. Female required more days than males for all macronutrients. Vitamin A and carotene were reported to require three weeks or more to estimate true intakes with adult females requiring over a month to estimate carotene. Results pertaining to vitamin C included 12 days for males and 7 days for females while vitamin E required 8 days for males and 16 days for females. In general, this study found that the population group that required the most days to estimate true intake was 5-17 y with adults requiring an intermediate amount (47).

Mennen et. al. reported analysis of the number of 24-hr DR required among French adults participating in the Supplémentation en Vitamines et Minéraux Antioxydants (SU.VI.MAX) Study which investigated the effects of antioxidant supplementation on cancer and heart disease (111). Participants



included in the additional analysis completed six 24-hr DR over a year and the study was separated into two phases consisting of 2 years each phase. Nutrients included energy and macronutrients with vitamin C, vitamin E, and  $\beta$ -carotene as the antioxidant micronutrients. Results from the first phase included the highest variance ratio for  $\beta$ -carotene and the lowest for carbohydrate. Carbohydrate required 5 days of DR while  $\beta$ -carotene required 16 recalls. For protein, total fat, and vitamin C, results showed 8 DR would be needed while vitamin E required 10 recalls for this French adult population. In general, the women required the same or more DR to estimate true intake for the macro- and micro-nutrients included (111).

A study was conducted in preschool age children reporting the variation in macronutrients and 11 micronutrient intakes stratified by age groups and gender (48). Huybrechts et. al. concluded that as the age of the children increased, the larger the variance ratios became and more day of DR were required for all nutrients. A 7-day DR would be sufficient to estimate energy and macronutrients when analyzing gender; however, results from the age groups indicates than more than 7 days would be required for the older children. Vitamin C could be estimated in 5 days among all age groups and genders (48). In an older adult population in Korea, the number of days to estimate energy, protein, fat, and carbohydrates among the males was over 2 weeks; however, vitamin C required 54 days to estimate true intake (97). The females required 8-23 days to estimate their macronutrient intake while vitamin C required 16 days. Oh et. al. concluded these results were attributed to the large within-person variability and low

between-person variability in this population group (97). Due to the population demographics and homogeneous population groups, many of these studies can only serve as implications for study design. There is limited data on nutrient variability and number of days need to assess nutrient intake, including antioxidants, among college age adults.

#### **2.4. Dietary Reference Intakes for Assessing Adequate Nutrient Intakes**

The essentiality of many macro- and micro-nutrients in preventing deficiencies requires a dietary recommendation for intake at a specified level in the general population. These recommendations are defined for many countries to assess nutrient intakes. For the U.S. and Canada, they are known as the Dietary Reference Intakes (DRIs) established by expert panels designated by the Institute of Medicine (IOM) (112). When adequate information is available, each nutrient is given specific DRIs which can include: an Estimated Average Requirement (EAR), a Recommended Dietary Allowances (RDA), an Adequate Intakes (AI) if the RDA is not available, and a Tolerable Upper Intake Limit (UL). With proper use of the appropriate DRI, these values provided for each specific age and life stage group are often used by government agencies in order to set standards for programs such as school meals or nutrient labeling on foods, by health professionals to provide counsel or interventions for individuals about dietary intake, and in health related research to assess the adequacy of usual nutrient intake among population groups (20). However, flavonoids and other polyphenolic compounds are still considered under review (21).

When assessing the adequacy of an individual's daily nutrient intake, many factors must be considered. It must be understood that nutrient intake alone, without biochemical and clinical data, is not sufficient to determine nutritional status. However, comparison of intake with a specific DRI is useful if usual nutrient intake can be estimated accurately (113). This can be a difficult process due to the fact that the truest intake requirement of an individual for a specific nutrient is not known and it is nearly impossible to measure habitual intake due to the sources of variation mentioned in earlier sections (45). It remains that the best estimate would be the midpoint of the distribution of requirements specific to their age and gender. This midpoint value is known as the EAR which defines the nutrient value to meet the requirement of 50% of healthy individuals. Any intake value above the EAR introduces the possibility that an individual may be consuming more than the requirement as well as any value below increases the possibility that they are not consuming adequate amounts. The RDA is set at 2 standard deviations away from the EAR and is the daily nutrient intake requirement to meet 97-98% of healthy individuals (20). For practical application of the DRIs, EAR values can be utilized with *individuals* and *population* groups to assess whether usual intakes are inadequate if it falls below the requirement. If an individual has usual intakes below the EAR, they increase the likelihood that their dietary intake needs improvement. Two methods can be used to apply the EAR to a population: the EAR cut-point method which includes the proportion of individuals in the group below the EAR, and the EAR probability approach which provides the probability of individuals in a group that their intakes

are inadequate (114). When utilizing the RDA and EAR for an individual, if the value falls below the RDA but above EAR, the probability of adequacy is below 97-98% and the diet quality might need improvement. Usual nutrient intakes of an individual at or above the RDA for a nutrient can be used to assess that the individual has low probability of being inadequate if long term diet records are used for assessment; however, the use of RDA values to determine inadequacy of a *population* group is inappropriate (88). It is suggested that using this approach will overestimate the prevalence of inadequacies compared to using the EAR (114).

Als for nutrient intake represent an estimate of mean intakes for a population that is apparently healthy and its use is limited as in comparison to the EAR and RDA. The AI is usually assumed to be higher than the RDA and therefore, intakes below the AI can not be assumed as inadequate (115). Individuals with intakes above the UL for specific nutrients can be assessed as at risk for adverse effects from excessive intakes. While proportions of individuals in a population above the UL may potentially be at risk for adverse effects, individual sensitivities to intake levels vary and use at a population level should be used with caution (116). For assessment of macronutrients, an Acceptable Macronutrient Distribution Range (AMDR) has been established. If an individual consumes average macronutrient intakes, as a percent of energy, within the establish AMDR, then it can be assumed that their diet is sufficient to reduce the risk of deficiency of the essential nutrients (117). While an individual below the specified DRI without biochemical data does not indicate that they are deficient,

the DRIs ensure the intake level is sufficient to meet the requirements of healthy individuals.

#### *2.4.1. Adequacy of Daily Nutrient Intake among Young Adults: Are They Meeting the Guidelines?*

Young adults today have had the benefit of growing up under the influence of dietary guidelines and national goals to improve health such as 5-A-Day campaign to increase fruit and vegetable intakes, Health Campus 2010, and MyPyramid (22, 23, 118). According to a recent review, 96% of U.S. young adults are considered to be in excellent or good health measured by traditional standards; however, in the same review, young adults ages 18-24 y had twice the mortality rate than adolescents ages 12-17 y (2). As with the general population, however, overweight/obesity has increased among adolescents and young adults which may increase risk factors for chronic diseases (119). The health and dietary intake status of young adults is a concern in the United States.

Recent studies have shown that many young adults are not meeting many of the established dietary guidelines or recommendations for adequate nutrient intakes and physical activity (25-27). In 2009, Burke et.al. conducted a risk screening initiative at the University of New Hampshire (27). They found that many of the male and female students had more adequate results concerning their macronutrient intake than micronutrient intake; however, this study utilized the RDA values to assess the micronutrient intake. For carbohydrate intake, 83% of the males and 82% of the females were within the AMDR for intake. Ninety-nine% and 98% of male and females met the protein guideline,

respectively. Fat intake had lower percentages meeting the guideline but the majority was within the adequate range (73% males and 69% females) (27). Overall, this college population group consumed macronutrient dense diets as evidences by the percentages meeting the guidelines.

The National Heart, Lung, and Blood Institute Growth and Health Study (NGHS) was designed to assess young African American and white females over the course of 10 years to evaluate obesity and their risk factors for chronic diseases such as cardiovascular disease (120). In 2007, Affenito et.al. further analyzed the micronutrient intake from the 3-day dietary records provided by NGHS (121). They included 1,166 white and 1,213 African American girls ages 9-18 y. Their results were produced using the EAR cut-point method and included adequacy assessment based on the age and gender specific EAR or AIs for each micronutrient. For vitamin A intakes, fewer than 54% of the girls had intakes below the guideline at all years. Most girls (81.2% or greater) had intakes below the guideline for vitamin E; however, percentage was dependent on age and race with the average intakes decreasing more among the white girls as age increased. Regardless of race, vitamin D and vitamin C intakes had an inverse relationship with age. More white girls than African American girls consumed adequate amounts of folate, however, in all years 46% of girls had intakes below the requirement. Vitamin B<sub>6</sub> and B<sub>12</sub> were generally consumed in adequate amounts with only 20.2% and 17.9% below the guideline, respectively. Calcium and magnesium consumption decreased across the years regardless of race. Finally, zinc consumption was adequate across the years with an increase

as age increased for white girls. Improvement of diet quality is greatly needed among this population group. While the authors do conclude that deficiencies in many of these nutrients are rare in the United States, these results could suggest that this young population is not consuming adequate fruits and vegetables to reduce the risk of chronic diseases. (121).

In another large scale study, known as the CSFII conducted by the USDA, nutrient intake adequacy was assessed using EARs for various adolescent age groups (122). Among the older adolescents, males and females ages 14-18 y, more than 50% consumed below the EAR for vitamin E, folate, and magnesium. In addition, more than 20% of the females in this age group were below the EAR for vitamins A and C, and zinc. Authors emphasized the importance of nutritional interventions for females aged 14-18 y (122). While adolescent populations are slightly younger than most college students or young adults, these studies report a decreasing trend for adequate intake as age increased which may put many of them at risk for insufficiency when entering young adulthood (121, 122).

Anding et. al. reported that majority of female college students were not meeting the 1995 Dietary Guidelines for Americans (DGA) in a subset population at Texas University (26). The 7 parameters outlined in the DGA include: eating a variety of foods, balancing the food eaten with physical activity to maintain or improve weight status, choosing a diet with plenty of grains, fruits, and vegetables, choosing a diet low in fat, saturated fat, and cholesterol, choosing a diet moderate in sugars, choosing a diet moderate in salt and sodium, and finally, drinking alcohol in a moderation. When the guideline pertaining to alcohol intake

was removed from the results, only 43% met at least one guideline and none of the participants complied with all 7 guidelines (26). These results could suggest that this population may not be aware of the guidelines or may lack the ability to make these guidelines a daily habit. In 2000, Lowry et.al. reported results from a questionnaire analyzing physical activity, food choices, and weight management goals among over 4,000 college students (30). They found that only 26.1% of the population was consuming 5 or more servings of fruits and vegetables with similar percentages among males and females. However, 78% of the population was consuming 2 or less servings of high fat foods. Servings of the recommended food groups below the guidelines can increase the likelihood that these young adults are consuming inadequate intakes of important micronutrients. (30). In general, conclusions from these studies suggest that college students overall are not meeting guidelines established to promote a healthy lifestyle that can reduce risks for chronic diseases.

#### *2.4.2. Lifestyle Factors Associated with Nutrient Intake among College Students*

For many individuals, this transition period of young adulthood includes establishing independence and this often involves making lifestyle decisions that impact their overall health and well being (123). Poor dietary intake and quality in conjunction with lack of physical activity are important contributors to the increasing rates of many health disparities that have affected every age group in the U.S. population (119, 124). Data shows that place of residence, new academic and social pressures, weight concerns, skipping meals, and access to fast food are a few contributing factors to inadequate nutrient intakes (24, 28-30).



In a recent study, Greaney et.al. identified factors that functioned as enablers or barriers to health in a subset population of college students (123). They reported that being physically active, regulating food intake, social support, healthy dining options at university dining services, and university environment to support physical activity as enablers to healthful behaviors. Barriers included high stress, time constraint, monetary cost of healthy foods, ready access to fast foods, and certain social situations (123). In a cross cultural study that analyzed physical activity levels, 58% of normal weight U.S. young adults participated in vigorous activity regularly and, in addition, more males than females were vigorously active (125). This is in contrast to the 37.6% reported among undergraduate students in a study by Lowry et. al. (30).

While the college environment often facilitates great social opportunities and support systems, it can also be a time of body transitions and weight concerns, especially for women (126-128). In an analysis of dietary behaviors in college students as related to dieting status, gender, and psychosocial variables, Leibman et.al. reported that more young adult females than males tend to diet, have lower self esteem, skip meals to lose weight, and have body dissatisfaction (95). These behaviors were documented as having a higher association with fat avoidance and disordered eating which can alter diet quality (95). While these issues are more prevalent among females, it should be addressed in both genders and its effects on nutrient adequacy and the accuracy of self reported intake data.

## **2.5. Supplement Use among U.S. Population Groups**

The use of dietary supplements in the U.S. among all age groups is becoming more prevalent and is continuing to increase (36). Data from NHANES has been used over the past 30 years to document supplement use in the U.S. (25). According to a recent analysis of the NHANES 2003-2006 data, supplement use was reported by 49% of the whole population over the age of 1 y and 54% among adults over the age of 20 y. Among adults, 56% of normal weight individuals reported supplement use. Supplementation was more prevalent among those individuals with higher education (61%) and among non-Hispanic whites (59%) (129). In general, the use of dietary supplements tends to increase with age with the highest prevalence among adults over 50 y of age and more common among women, those who are more physically active, have a higher income, and consume a more micronutrient dense diet (130). Overall, supplement use among the U.S. population has increased approximately 10% since NHANES III 1988-1994 (129).

While supplement use has increased when analyzing the U.S. population as a whole, adolescent supplement intake has remained consistent around 29% of the population according to a report of the dietary trends in the U.S. (27). Results from the NHANES 1999-2004 data indicate that 34% of children and adolescents report vitamin and mineral supplement use (55). In another study with adolescents in 2008, 71% of the total 3,428 students who completed a self reported survey documented supplement use (131). In a study performed in university students analyzing the use of non-vitamin and non-mineral

supplements, 26% of the 263 student participants reported use of a supplement (57). However, there is a gap in the literature reporting general supplement use among college students or young adults; more studies analyze specific supplement use (132-136) or supplementation in a certain subset population of young adults (137-141).

#### *2.5.1. Supplement Type and Frequency*

While supplement use is increasing across the U.S. population, it is suggested that only individuals who restrict energy intake or are on a severe weight loss diet, eliminate a food group from usual diet, or who consume high carbohydrate and low micronutrient dense diets should use supplements (36). Results from NHANES 2003-2006 also documented that the majority of the population reported the use of only one supplement with multi-vitamin/multi-mineral being the most prevalent (33%) followed by botanical supplements and amino acids. The lowest prevalence of a vitamin supplement was reported among adolescents ages 14-18 y; among females, the highest prevalence of an iron supplement was ages 19-30 y and 31-50 y. In addition among supplement users, 79% report daily use over the past 30 days (129).

Perkins et.al. additionally reported that the most common non-vitamin and non-mineral supplements among college students were ginseng, Echinacea, and protein powders/amino (57). Another study reports that adolescent males document greater use of ergogenic aids such protein supplements while females more frequently consume herbal supplements related to weight loss (142).

### *2.5.2. Contribution of Supplement to Nutrient Adequacy*

The ADA position on supplementation states that daily nutrient intake from a wide variety of foods is recommended to meet adequate requirements over the use of a supplement (24). However, as previously established, adequacy from dietary sources only is low for many nutrients among individuals in this population group (27, 121, 143). Low intakes of many vital nutrients can put individuals at risk for deficiencies and their consequences. Low intakes of calcium and vitamin D, specifically for females, increases their risk of poor bone health in the future (144). Folate intakes in this female population is of great importance due to being of childbearing age and the prevention of neural tube defects (145). Low iron intake from dietary sources is common among young females due to overall lower calorie intake or poor intake of heme iron sources from animal products and can increase their risk for iron-deficient anemia (146). Supplementation may be suggested by health professionals in certain individuals with habitual low dietary intakes (24).

In 2000, Stang et.al. reported supplement use and dietary adequacy among an adolescent population (147). They utilized data from the 1994 CSFII. Out of the total 423 adolescents included, one third was classified as supplement users and 15.6% reported daily use. Multivitamin was the most common with 65.5% of users reporting consumption. Participants who were non-users consumed a greater percentage of energy from total fat and saturated fat but less from carbohydrate than users. Users had higher micronutrient intake than non-users, except for zinc. One third of the adolescent males in all categories of

supplement use consumed less than 75% of the RDA for vitamins A and E, calcium, and zinc. More than one fourth of the male non-users consumed below 75% of the RDA for vitamins B<sub>6</sub> and C. For the females, 37% consumed 75% below the RDA for vitamins A and E, calcium, iron, and zinc in all categories of supplement use. Among non-users, 35% of the females consumed below the RDA for vitamins B<sub>6</sub> and C, and folic acid (147). Unfortunately, Stang et.al. also reported that adolescents that had low nutrient intakes were less likely to take a supplement. Therefore, primary prevention programs should be established targeting the nutrient quality in young adult population groups (147).

In an adult population, Archer et.al. documented considerable benefit for many micronutrients with the addition of a supplement to daily food intake (148). Dietary intake data was collected from the International Study of Macro- and Micro-nutrients And Blood Pressure (INTERMAP) and nutrient adequacy from diet and supplement was compared to the EAR or AI for nutrient intake. In total, they reported that average intake from foods for vitamins A and C, and niacin exceeded the EAR but not vitamin E and folate. Supplement users had higher intakes from food for vitamin C and folate than non-user. With the addition of a supplement, users had higher intakes than non-users for vitamins A, C, and E, niacin, and folate. Total intakes among the supplement users for selenium, zinc, phosphorus, magnesium, vitamins C and E, niacin, and folate were considerably above 100% of the EARs but non-users were below the EAR for vitamin E and folate (148). Among a Canadian adult population 19 y and older, supplementation increased adequacy for many nutrients (149). Participants who

consumed a multivitamin had average intakes from diet and supplement of folate above the RDA and, among the women 19-50 y, iron intakes increased above the RDA after supplementation. From dietary sources only, supplement users had similar intakes as non-users for iron, calcium, and folate with many of the intake below the RDA/AI for females ages 19-35 y. However, with the addition of a supplement, calcium and vitamin D intakes increased above the recommendations among all gender and age groups (149). In conclusion, supplementation sizably increased the micronutrient intakes among this adult population.

While supplementation may play a significant role in overall nutrient adequacy (35), some may contain higher amounts than needed which could increase risk of toxicity or may contain compounds that do not have an established requirement (150). Most nutrients consumed at or above the UL from dietary sources only have not been shown to have adverse effects. The issue arises with supplemental forms or fortification of foods (36). Supplements are over a 25 billion dollar industry in the United States and many remain unregulated (151). The naivety and often unadvised usage of this young adult population group puts them at risk for adverse side effects of over supplementation (152) and should be addressed by health professionals.

In conclusion, estimation of usual nutrient intake, including antioxidant nutrients, from dietary sources and supplement use among college aged adults is a vital part in assessing dietary quality and the risk of developing diseases. In

order to do so, nutrient intake data must be validated by identifying misreporters and the variation of nutrient intakes among this population must be controlled. While access to healthy foods and means for physical activity in a college environment is available, many young adults are not meeting the guidelines for nutrient intake or physical activity which increases risk factors for deficiency and disease. While research is often devoted to assessing diet quality and disease risk factors in adults, there is limited data in comparison that addresses this influential population.

Table 1. A comparison of micronutrient intakes by gender and age

Nutrient	Agriculture Research Service, 2007-2008 (101)		Burke, 2009 (27)	Lambert, 2004 (102)	Kimura, 2003 (103)
	12-20 y	20-29 y	18-24 y	15-18 y	18 y
Vitamin A, µg RAE					
Male	680±47.1	597±28.6	2,666.2±2,135.6	-	-
Female	528±33.5	532±32.7	2,399.2±1,776.3	-	705±435
Alpha-carotene, µg					
Male	252±41.9	238±36.9	-	-	-
Female	242±50.3	274±33.2	-	-	-
Beta-carotene, µg					
Male	1,368±183.7	1,452±147.8	-	-	-
Female	1,114±111.0	1,606±226.5	-	-	-
Beta-cryptoxanthin, µg					
Male	68±8.3	69±7.3	-	-	-
Female	56±8.8	76±18.1	-	-	-
Lycopene, µg					
Male	6,708±748.2	7,886±988.8	-	-	-
Female	4,265±491.5	5,219±804.6	-	-	-
Lutein + Zeaxanthin, µg					
Male	1,082±203.3	1,022±109.6	-	-	-
Female	740±103.5	1,362±209.7	-	-	-
Thiamin, mg					
Male	1.88±0.061	2.18±0.187	-	1.4-1.8	-
Female	1.45±0.091	1.38±0.044	-	1.2-1.5	0.7±0.3
Riboflavin, mg					
Male	2.58±0.105	2.60±0.139	-	1.6-2.3	-
Female	1.78±0.074	1.81±0.099	-	1.3-1.8	1.1±0.4
Niacin, mg					
Male	28.9±1.34	34.2±1.61	-	30-40	-
Female	20.8±0.77	21.0±0.68	-	23-27	23± 7
Vitamin B <sub>6</sub> , mg					
Male	2.29±0.132	2.57±0.140	-	1.6-2.2	-
Female	1.63±0.059	1.66±0.089	-	1.4-1.6	0.9 ±0.4
Folate, µg DFE					
Male	610±15.9	692±39.5	434.0±301.6	-	-
Female	509±32.6	460±28.1	383.0±325.2	-	-
Vitamin B <sub>12</sub> , µg					
Male	6.68±0.277	6.95±0.382	-	5.0-7.0	-
Female	4.14±0.227	4.17±0.226	-	3.4-5.0	4.4±4.1
Vitamin C, mg					
Male	86.6±5.67	93.1±7.08	172.7±150.6	70-100	-
Female	73.8±5.64	80.8±8.49	128.6±97.9	70-100	73±38
Vitamin D, µg					
Male	5.9±0.44	4.9±0.21	5.7±4.9	1.8-6.5	-
Female	3.8±0.20	3.6±0.25	3.7±3.1	1.4-4.6	6±8



Vitamin E, mg $\alpha$ -tocopherol					
Male	7.7 $\pm$ 0.49	7.9 $\pm$ 0.27	-	3.2-32.4	-
Female	6.0 $\pm$ 0.35	6.5 $\pm$ 0.47	-	3.2-32.4	7.7 $\pm$ 3.0
Calcium, mg					
Male	1,173 $\pm$ 52.8	1,150 $\pm$ 52.7	1,206.6 $\pm$ 547.0	500-1,200	-
Female	878 $\pm$ 40.8	869 $\pm$ 36.5	904.1 $\pm$ 430.3	500-1,200	-
Magnesium, mg					
Male	282 $\pm$ 10.8	336 $\pm$ 12.1	-	350-375	-
Female	223 $\pm$ 9.7	246 $\pm$ 11.7	-	250-275	-
Iron, mg					
Male	16.6 $\pm$ 0.40	18.1 $\pm$ 0.73	20.4 $\pm$ 8.73	-	-
Female	13.8 $\pm$ 0.80	12.6 $\pm$ 0.42	15.2 $\pm$ 6.7	-	-
Zinc, mg					
Male	13.2 $\pm$ 0.36	15.2 $\pm$ 0.69	-	-	-
Female	9.6 $\pm$ 0.46	9.7 $\pm$ 0.24	-	-	-
Selenium, $\mu$ g					
Male	125.2 $\pm$ 4.36	143.1 $\pm$ 3.79	-	-	-
Female	88.3 $\pm$ 3.78	90.0 $\pm$ 2.35	-	-	-
Potassium, mg					
Male	2,587 $\pm$ 108.4	2,939 $\pm$ 117.3	3,345.2 $\pm$ 1,578.3	3,200-3,800	-
Female	1,957 $\pm$ 54.2	2,094 $\pm$ 67.5	2,449.2 $\pm$ 1,169.4	2,200-3,000	-

## **Chapter 3**

### **Materials and Methods**

#### **3.1. Study Design and Population**

Participants were recruited from the University of Connecticut (UCONN) located in Storrs, Connecticut. Initial recruitment began in introductory nutrition courses and eventually expanded to large classrooms in all programs. Flyers were posted in the Student Union, dormitories, and buildings throughout the Storrs campus. Campus wide emails were sent out with study purpose and eligibility requirements. The study and all recruitment materials were approved by the UCONN Institutional Review Board for inclusion of human participants.

This study recruited males and females between the ages of 18-25 y who were apparently healthy. Participants were excluded if taking any prescribed medication or history of chronic disease. All forms of supplements were included. All visits took place in Dr. Chun's laboratory at the Storrs campus. At the initial visit, participants signed a consent form, answered eligibility requirements pertaining to medical history, and were informed of their responsibilities over the course of the study. Height and weight were measured and BMI was calculated as  $\text{kg/m}^2$ . Blood pressure was measured twice while remaining seated and a fasting finger stick blood sample was collected to measure lipid profile (Cholestech, LDX, Hayward CA). If BMI, blood pressure, and lipid profile measurements were within a healthy range (153), participants were included. The study recruited 77 eligible participants with a 22% drop out

rate. This study retained 60 participants for the month of study including 40 females and 20 males.

### **3.2 Dietary Assessment**

Each eligible participant was asked to fill out a Health and Nutrition Survey which included but not limited to personal information about race, class standing, major, residence, meal plan consumption, physical activity, body image perception, and if currently using any form of dietary supplement. In addition, they were given detailed instructions by an experienced research staff on how to complete a 24-h DR that was to be collected at the end of every day for 30 consecutive days. The use of 30 days of dietary assessment was to decrease the within-person variation bias notoriously associated with sampling a fewer number of daily intakes specifically for the nutrients that are less frequently consumed (41). An experienced research staff was used to collect and enter dietary information in order to limit observer bias. Only participants who completed the total 30 consecutive DR were included in dietary analysis. Each DR included information on supplement intake, brand name, type of nutrient, and dosage. The DR were emailed to password protected email account and only accessed by the research staff. If any incomplete DRs were emailed or participants missed days, the individual was promptly contacted for further explanation and detail. In data analysis, supplement users were defined as consuming one or more dietary supplements more than once per week. A supplement was defined according to the Dietary Supplement Health and

Education Act (DSHEA) of 1994 (154). In brief, a supplement is a product that intends to supplement the diet and can contain any of the following ingredients: a vitamin, a mineral, an herb or other botanical, an amino acid, or a concentrate, metabolite, a constituent, extract, or combination of any ingredient. A supplement must be identified as a dietary supplement on the label and must not be represented as the only component of a meal or diet (154).

### **3.3. Nutrient Analysis**

Dietary intake data were collected and analyzed using Nutrition Data System for Research (NDSR) software version 2009, developed by the Nutrition Coordinating Center (NCC), University of Minnesota, Minneapolis, MN. NDSR provides a complete nutrient profile for all foods in the database, excluding flavonoids and proanthocyanidins through output in an excel file. In order to calculate the lacking nutrients, the NCC Flavonoid and Proanthocyanidin Provisional Table was used. The NCC table links foods from the NDSR output file with flavonoid and proanthocyanidin values provided in the USDA Special Interest databases. The USDA Database for the Flavonoid Content of Selected Foods, Release 2.1 (January 2007) is the major source for the values in this table. The NCC table provides values for 33 flavonoids and 6 classes of proanthocyanidins. The NDSR output file and the NCC table were linked through Statistical Analysis Systems (SAS) statistical software package version 9.2 (SAS Institute, Cary, NC, USA).

Nutrients included in analysis from the NDSR output and NCC table for objectives 1 and 2 include: total energy (kcal), total fat (g), total carbohydrates (g), total protein (g); vitamin A ( $\mu\text{g}$  RAE, thiamin (mg), riboflavin (mg), niacin (mg), vitamin B<sub>6</sub> (mg), folate ( $\mu\text{g}$  DFE), vitamin B<sub>12</sub> ( $\mu\text{g}$ ), vitamin D ( $\mu\text{g}$ ), calcium (mg), iron (mg), magnesium (mg), selenium (mg), zinc (mg), and antioxidant nutrients  $\alpha$ -tocopherol (mg),  $\gamma$ - tocopherol (mg), and total vitamin E (mg) ( $\alpha$ -, $\beta$ -, $\delta$ -, $\gamma$ - tocopherol); vitamin C (mg),  $\alpha$ - carotene ( $\mu\text{g}$ ),  $\beta$ -carotene ( $\mu\text{g}$ ),  $\beta$ -cryptoxanthin ( $\mu\text{g}$ ), lutein + zeaxanthin ( $\mu\text{g}$ ), lycopene ( $\mu\text{g}$ ), and total carotenoids ( $\mu\text{g}$ ) ( $\alpha$ -, $\beta$ -carotene,  $\beta$ -cryptoxanthin, lutein, lycopene, and zeaxanthin); total flavonoids (mg) (flavonols, flavanones, flavan-3-ols, anthocyanidins, and isoflavones), isoflavones (mg) (daidzein, genistein, glycitein, coumestrol, biochanin A, and formononetin), and proanthocyanidins(mg) (monomers, dimers, trimers, 4-6 mers, 7-10 mers, and polymers).

The analysis from the NDSR output and NCC table for objective 3 include only nutrients with a defined DRI: total fat (g), total carbohydrates (g), total protein (g); vitamin A ( $\mu\text{g}$  RAE, thiamin (mg), riboflavin (mg), niacin (mg), vitamin B<sub>6</sub> (mg), folate ( $\mu\text{g}$  DFE), vitamin B<sub>12</sub> ( $\mu\text{g}$ ), vitamin C (mg), vitamin D ( $\mu\text{g}$ ), vitamin E (mg  $\alpha$ -tocopherol), calcium (mg), iron (mg), magnesium (mg), selenium (mg), zinc (mg), and total flavonoids (mg) (flavonols, flavanones, flavan-3-ols, anthocyanidins, and isoflavones) from supplements. Nutrients from supplements were analyzed through NDSR for each supplement user. However, if the specific supplement was not found in the program, details of ingredients and nutrition labels were searched for using the Internet. Each nutrient dose provided by a

supplement was multiplied by frequency of consumption and averaged over 30 days.

### 3.4. Misreporting nutrient intake

Due to the serious problem of misreporting in studies involving dietary assessment and the higher prevalence associated with the use of long term dietary records, exclusion of underreporters as well as overreporters of energy intake is strongly suggested (155). Energy intake ( $El_{rep}$ ) was averaged from the reported 30-day DR for each individual. Each subject's height and weight were measured at the first visit and applied values to the Schofield age and sex specific formulas (156) to measure the estimated basal metabolic rate ( $BMR_{est}$ ). These values were used to calculate  $El_{rep}:BMR_{est}$  ratio for each subject. Black et al. (15) re-evaluated the Goldberg cut-off equation previously used (91, 93) and provided guidelines for choosing an appropriate physical activity level (PAL) value for the study population. These investigators stated that the previously cited PAL of 1.55 in many studies should only be used with a population group that is sedentary (40). Therefore, a PAL value of 1.6 was chosen for this population according to the results from the questions pertaining to physical activity in the health and nutrition survey and the WHO/FAO recommendations for energy requirements (92). A value too high can increase the number of under reporters and exclude valuable data. PAL level of 1.6 was applied to the following equation (40):

$$El_{rep}:BMR_{est} > PAL \times \exp \left[ \frac{El_{rep} - BMR_{est}}{BMR_{est}} \right] \quad (1)$$

$$EI_{rep}:BMR_{est}<PAL \times \exp\left[\frac{s.d. \times z}{n}\right] \quad (2)$$

Where s.d.min is -2 for 95% the lower confidence limit and s.d. max is +2 for 95% upper confidence limit (CI),  $n$  was the number of participants, and  $S$  is calculated from (15):

$$S = \frac{1}{d} \left[ \frac{CV_{wEI}^2}{2} + \frac{CV_{wB}^2}{2} + \frac{CV_{tP}^2}{2} \right] \quad (3)$$

$S$  variable includes variation in intake, BMR, and energy requirements.  $CV_{wEI}$  is the within subject coefficient of variation in energy intake,  $d$  is the total number of days diet was recorded,  $CV_{wB}$  is the coefficient of variation of repeated BMR measurements or the precision of estimated compared with measured BMR, and  $CV_{tP}$  is the coefficient of variation derived from the mean and standard deviation of the study. In this study, the  $CV_{wEI}$  was calculated from the average reported energy intake for all 60 participants. The  $CV_{wB}$  and  $CV_{tP}$  values were derived by Black et. al. and deemed appropriate for application to future studies (40). Calculations (1) and (2) were applied to each subject with a PAL level set at 1.6 across gender using Microsoft Excel, version 2003. Any value for a given participant below calculation (1) was classified as underreporter and any value for a given participant above calculation (2) was classified as an overreporter. Only values in between these confidence intervals were included in the analyses of this present study.

### 3.5 Statistical Analysis

Data analysis for mean, within- and between-person variation, and the variance ratios were executed using SAS version 9.2 (SAS Institute, Cary, NC, USA). The accuracy of these estimations is dependent on the normality of the nutrient intake distributions for the population. Although data analysis using Kolmogorov's D-statistic for normality testing is not suggested for data that is not independent (157), it was used to evaluate the skewness from histograms and normal probability plots. All nutrient intakes for the population in this study were classified as non normal by a D-statistic  $>0.05$ . Therefore, all data was  $\log_e$  transformed and retested for normality. However, there are major limitations with the use of transformed data in relation to back transformation to original scale. Estimations based on transformed nutrient data are difficult to interpret and provide valid conclusions as well naïve back transformation can introduce immeasurably bias (99). Authors of previous studies have reported results using varying methods with the transformed data despite the bias associated with the back-transformation methods (47, 158) while others have reported untransformed data due to the fact that the transformation did not considerably alter the variance components (97, 99). While an approach has been describe to remove the back transformation bias associated with estimates of usual intake (41), there is no method concerning variance estimates. Therefore, all results in objective 1 and 2 are untransformed.



The dependent variables in this model were energy intake and 30 nutrients. Nutrients from supplements were not included in these analyses. A mixed effects regression model with a restricted likelihood estimator was used to estimate mean intake, standard deviation, and the within-person variation ( $S_w$ ) and between-person variation ( $S_b$ ) among males, females, and total population. The variation components are the square root of the estimated within-person variation ( $S_w^2$ ) and the between-person variation variance ( $S_b^2$ ). The variance ratios of within- and between-person variation were expressed as  $S_w^2 / S_b^2$ . The coefficient of variation (CVs) within and between were calculated as:  $CV_w = [S_w / \text{mean intake (nutrient)}] \times 100$ ; and  $CV_b = [S_b / \text{mean intake (nutrient)}] \times 100$ . In statistical models from previous studies, fixed effects or sometimes termed as nuisance effects of age, ethnicity or race, income or education level, day of the week, sequence of interview, and dietary assessment site were controlled to reduce the within- and between-person variation (41, 99, 121, 159). However, due to the homogeneity of the sample population in this study as well as the use of 30 consecutive days, many of these predictors do not apply. Therefore, the statistical models in this present study controlled for the random effects of the subject ID and the fixed effects of energy intake. In addition, gender was controlled for when estimates were presented for the total population. Nutrient intake values in text are mean  $\pm$  standard deviation (SD).

For objective 2, usual nutrient intake distribution graphs for the total population and the calculation of number of days required to correctly classify individuals with a given level of accuracy were produced using SAS, version 9.2.

Microsoft Excel, version 2003 was used to randomly select days from the total 30 for each subject. The day that was randomly selected was the single day average intake. In addition, it marked the first day out the seven consecutive days chosen for a separate calculation of average intake. If the day that was randomly selected was near the end of the recording period and therefore not able to choose the following 6 days, the additional days needed were selected from the days preceding the random first day in order to represent each day of the week. The calculation of  $D$  is also dependent on a hypothetical correlation coefficient ( $r$ ) between the observed and the true intakes. As  $r$  increases, the percentage of participants correctly classified increases and the misclassification decreases. Therefore,  $r$  was set at 0.9 in order for 90% confidence that 80% of the participants are accurately classified into thirds of a distribution and less than 1% is misclassified according to previous published work (47, 110). The number of days ( $D$ ) was calculated using the following formula (26):

$$D = \left\lceil \frac{1}{r^2} \right\rceil \quad (4)$$

The untransformed within- and between-person variation estimated from objective 1 were included in this calculation expressed as the variance ratio ( $S^2_w / S^2_b$ ). The smaller the within-person variation compared to the between-person variation and the ratio as a whole, the fewer amount of days is expected to be required for each nutrient. This analysis was performed including all participants and stratified by gender.

The EAR cut-point method includes calculating the individuals who have usual nutrient intakes below the EAR for a specific nutrient requirement defined

by the IOM (20, 49). Use of this method is based on the following assumptions: 1) intakes and requirements are not correlated (all nutrients satisfy this assumption except energy), 2) the distribution of requirements is symmetrical. This is assumed for all nutrients except iron, particularly among women in reproductive years due to blood and iron losses during menstruation. Therefore, the EAR cut-point method cannot be used to assess inadequacy from iron intakes and iron was not included in this analysis. And 3) the distribution of intakes is more variable than the distribution of requirements (114). While this present study was developed under the assumptions that using 30 days of dietary assessment would greatly decrease the within-person variation to approach values near the between-person variation in order to provide more representative values for usual nutrient intake, the calculation of the accurate number of days of dietary assessment to estimate usual nutrient intake was performed as a preliminary step to this present objective. The estimation of the prevalence of inadequacy is dependent on usual nutrient intakes and intakes that are normally distributed (41). Therefore, the mean intakes from diet and supplement included in this analysis were  $\log_e$  transformed to normality and then back transformed using natural log to original scale. All nutrient intake data was analyzed using a mixed effects regression model with a restricted likelihood estimator and adjusted for the random effects of subject ID and the fixed effects of energy intake. Analysis of supplement intake included additional adjustment of gender. The results were then used to estimate nutrient intake adequacy by applying the EAR cut-point method to determine the proportion (%) of individuals

with usual intakes from diet and supplement below the EAR. Percent of energy from fat, carbohydrates, and protein were calculated by multiplying the total mean intake of the macronutrient (g) by its energy density (9 kcal/g fat or 4 kcal/g carbohydrate and protein, respectively), then dividing the result by the total energy intake for each participant. Chi-squared analysis was used to determine differences between the number of male and female supplement users who used each form of supplement. The criterion for statistical significance was at P value < 0.05.

## Chapter 4

### Results

#### 4.1. Misreporting and Usual Intake for Energy and Key Nutrients among College Students

##### 4.1.1. Under- and Over-reporting

**Table 4.1** reports the results from the original sample of college students and misreporting of energy intake. The original sample was comprised of 20 males and 40 females with the mean age of 20 y. Anthropometric results displayed that the mean BMI values for males was  $23.9 \pm 3.1$  and  $22.7 \pm 2.4$  for the females which is within a healthy range. While the standard deviations would classify some of the participants as overweight, the results are likely due to muscle weight and not contributed to fat composition. The mean EI:BMR<sub>est</sub> ratio for males and females were 1.4 and 1.3, respectively. After application of the equation, the cutoff limits for the population in this study were less than 1.12 were considered underreporters and over 2.28 were classified as overreporters of energy intake. Overall, 27% of the population, 15% of males and 30% of females, were identified as underreporters when individualized EI:BMR<sub>est</sub> ratios were applied to the confidence intervals. While no males were classified as overreporters, 2.5% of females exceeded the cutoff limit. Therefore, a total of 44 participants, 17 males and 27 females, were classified as average reporters and included in dietary assessment analysis.

#### *4.1.2. Lifestyle Characteristics of Participants who were Average Reporters*

**Table 4.2** reports lifestyle characteristics of the remaining participants who were classified as average reporters. The majority of the remaining participants were non-Hispanic white for both males and females (82% and 74%, respectively). Each year of study was represented by both genders, except graduate level, with the vast majority being in their freshman or sophomore year. Overall, the study population participates in daily physical activity at or above the recommended duration and intensity defined by the American College of Sports Medicine (ACSM) and American Heart Association (AHA) for adults (160). More females than males reported participating in greater than 30 minutes per day of moderate activity which included activities such as brisk walking and bicycling. However, 70% of the females and 88% of the males reported greater than 30 minutes per day of vigorous activity which included heavy aerobic exercise or activity that increases heart rate. Only 18% of males and 48% of females had a declared major involving health sciences. Approximately 65% of the males and 78% of the females reported that they had a campus meal plan and consumed their meals in the dining halls or cafés located throughout campus. Less than half of the participants reported consuming fast food for both genders (24% of males and 37% of females). Concerns and behaviors involving weight issues were more prevalent among the female participants with 63% of females reporting feelings of pressure to be a certain weight. However, only 37% and 30% of females reported a moderate to severe fear of gaining weight and skipping meals to lose weight, respectively.

#### *4.1.3. Mean Intakes and Variance Components*

**Figure 4.1** compares the daily variation of total fat intake (g) and total flavonoid intake (mg) from three participants, one in the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of the 30-day intakes. There was a higher degree of variation between individuals in the 90<sup>th</sup> percentile than that of the subject in the 10<sup>th</sup> percentile for total flavonoid intake when compared to total fat intake. Due to the high degree of within-person variation expected in dietary assessments of general population, any individual's daily consumption of a nutrient may fall into other percentiles but the overall averages differ largely. The participant in the 90<sup>th</sup> percentile for total fat consumed 3 times more than the participant in the 10<sup>th</sup> percentile and the 90<sup>th</sup> percentile participant consumed 18 times more total flavonoids than the participant found in the 10<sup>th</sup> percentile.

**Table 4.3** displays the means, variance components, and the number of days for energy and select nutrients, including antioxidants, for 44 college students. For all nutrients except protein, riboflavin, vitamin B<sub>6</sub>, and magnesium after adjustment for gender only, the within-person coefficient of variation is larger than the between-person coefficient of variation. This resulted in variance ratios <1 for those select nutrients. Among the micronutrients, the within-person coefficient of variation differed more markedly and was more than 2 times higher than the between-person coefficient of variation for  $\alpha$ -carotene,  $\beta$ -cryptoxanthin, lutein+zeaxanthin, vitamin B<sub>12</sub>,  $\gamma$ -tocopherol, and isoflavones which resulted in large variance ratios ranging from 4.23-10.56. After adjusting for energy intake, the within-person coefficient of variation approached the between-person

coefficient of variation for only fat, carbohydrates,  $\gamma$ -tocopherol, and proanthocyanidins. For the remaining nutrients, the variance components remained the same or increased slightly.

**Table 4.4** presents the results from mean intakes, variance components, and number of days among male and female college students. Males had significantly higher mean intakes for all nutrients excluding all carotenoids, vitamin C,  $\gamma$ -tocopherol, flavonoids, isoflavones, and proanthocyanidins (P value <0.05). However, after adjusting for energy, males consumed higher intakes for only 15 of the 31 nutrients including protein, vitamin A, all B-vitamins (excluding thiamin), vitamin D,  $\alpha$ -tocopherol, iron, magnesium, selenium, zinc and flavonoids (P value <0.05).

Compared to the males, the females had lower coefficient of variation within and between for all nutrients except total flavonoids and isoflavones. Females also had higher values for the between-person variation for lutein + zeaxanthin and  $\gamma$ -tocopherol but higher values for the within-person variation for lycopene, total carotenoids, selenium, and proanthocyanidins. However the variance ratios for females, with the exception of  $\gamma$ -tocopherol, total flavonoids, lutein + zeaxanthin and  $\beta$ -cryptoxanthin, are larger and more variable than the males. Variance ratios were generally >1 for macro- and micronutrients for both sexes with the exception of protein, lycopene, riboflavin, niacin, vitamin B<sub>6</sub>, folate, calcium, and magnesium for the males. Overall, females had lower day-to-day variability among most nutrients compared to males, however, the small between-person variation resulted in larger variance ratios among the females.



The larger between-person coefficients of variation for the males reflect the heterogeneity of this population.

After adjusting for energy intake in order to decrease the population variability, the coefficient of variation for within- and between-participants decreased for both genders for all macronutrients but results for the micronutrients were more variable. For both genders, the within- and between-person coefficient of variation was decreased for vitamin A, all B-vitamins, vitamin D,  $\alpha$ -tocopherol, total vitamin E, calcium, iron, magnesium, selenium, and zinc. However, this subsequently increased the variance ratios for these nutrients for both genders indicating that the between-person coefficient of variation was decreased to a greater magnitude among the genders. For all the carotenoids, flavonoids, isoflavones, and proanthocyanidins, the between-person coefficient of variation slightly increased for both genders which decreased the variance ratios for those nutrients. For the males, the within-person coefficient of variation was decreased but the between-person coefficient of variation increased for  $\gamma$ -tocopherol which markedly decreased the variance ratio. However, for the females, both the within- and between-person coefficient of variation was decreased for  $\gamma$ -tocopherol which increased the variance ratio. Overall, adjusting for energy reduced the within- and between-person variation for 21 of the 31 nutrients for both males and females in this population group; however, the magnitude of the reduction in the coefficients of variation was similar between genders indicating that adjusting for energy did not considerably weaken the differences between genders.

## **4.2. The Number of Days to Accurately Assess Usual Intakes for Energy and Key Nutrients, Including Antioxidants, among College Students**

### *4.2.1. Effects of the Number of Dietary Recalls on Intake Distributions*

The effects of randomly selecting smaller amount of DR days from the total 30 days to represent shorter recording periods most frequently used in nutritional studies are shown in **Figure 4.2**. The graphs display between-person variation as the within-person variation is canceled out due to the large number of days included. Nutrients included in this analysis were energy, vitamin C, total flavonoid, and isoflavone intakes. These nutrient distributions were compared to the total intake in one month. As the number of days of dietary assessment increased, the standard deviation between participants decreased and the distributions became more normalized for vitamin C, total flavonoid, and isoflavones. However, energy intake distribution became more skewed when comparing 7 days to 30 days despite decrease in SD. When analyzing data based on only one day for each participant for total energy intake, the 90<sup>th</sup> percentile consumers were 3 greater than those consuming intakes in the 10<sup>th</sup> percentile (**Figure 4.2-1**). The effect was even greater in the micronutrients. At the 90<sup>th</sup> percentile for intake of vitamin C, participants consumed 23 times more than those in the 10<sup>th</sup> percentile (**Figure 4.2-2**). The 90<sup>th</sup> percentile intakes for total flavonoids and isoflavones are 285 and 2,044 times greater than that of the 10<sup>th</sup> percentile participants, respectively (**Figures 4.2-3 and 4.2-4**). This decreasing trend continued for data from the seven days with ratios of 2.4 for energy, 4.3 for vitamin C, 45.9 for total flavonoid, and 133.8 for isoflavones. The

ratios for are less dramatic when the days were increased to the total 30 for energy and vitamin C with differences of 2.2 and 4.0, respectively. However, the polyphenols intake had a much greater decrease with ratios of 27.8 for total flavonoid and 60.9 for isoflavones.

#### *4.2.2. Number of Days of Dietary Assessment Required to Assess Usual Antioxidant Intake*

In addition, results from the calculation of days for each nutrient required to achieve  $r \geq 0.9$  for total population and by gender can be seen in **Figure 4.3**. After adjusting for energy intake and gender for the nutrient intakes of the total population, fat, carbohydrate, protein,  $\alpha$ -tocopherol, lycopene, and proanthocyanidins intakes could be estimated with 7 or fewer days of dietary assessment.  $\beta$ -carotene, vitamin C, total carotenoids, and flavonoids intakes could be assessed with 14 days or two sets of 7 day-DRs. Overall, 30 days is sufficient to estimate usual nutrient intakes for this total population group (excluding  $\beta$ -cryptoxanthin).

Among the males, the majority of the nutrients could be assessed with 7 days or fewer excluding energy, fat, all carotenoids (except lycopene and total carotenoids), vitamin C,  $\gamma$ -tocopherol, total vitamin E, flavonoids, and isoflavones after adjusting for energy intake (**Figure 4.3**). Among these nutrients requiring more than 7 days of dietary assessment, only energy, fat, vitamin C,  $\beta$ -carotene, and flavonoids could be assessed accurately within 14 days or two sets of 7-day DRs. Compared to the males, females required more days to assess nutrient intakes except for lutein + zeaxanthin and flavonoid intakes (**Figure 4.3**). Only

flavonoid intakes could be assessed with 7 days or fewer of dietary assessment among females after adjusting for energy intake. Among the remaining nutrients, energy, carbohydrates, fat, vitamin C, lutein + zeaxanthin, and  $\beta$ -carotene intakes could be assessed within 14 days or two sets of 7-day DRs. The remaining nutrients would require more than 14 days of dietary assessment. Overall, 30 days was sufficient to estimate usual nutrient intakes for both males and females for the majority of the nutrients (excluding  $\beta$ -cryptoxanthin, lutein + zeaxanthin and  $\gamma$ -tocopherol for males, and lycopene for females).

### **4.3. Usual Nutrient Intake Adequacy from Diet and from Supplements**

#### **Assessed by the EAR Cut-Point Method among College Students**

##### *4.3.1. Nutrient intake adequacy among genders*

**Table 4.5** presents the results from the calculation of days of dietary assessment for the remaining nutrients with a defined EAR. All nutrient intakes for both males and females could be estimated within 30 days. Therefore, 30 days was sufficient to represent usual nutrient intakes and the EAR cut-point method could be used for the mean intake over 30 days for each individual to determine adequacy. **Table 4.6** presents the mean intakes, % of energy from macronutrients, and proportion (%) of individuals within the AMDR and below the EAR for select nutrients by gender. The percentages of energy coming from macronutrient intakes were similar among the genders with males consuming slightly higher percentages of fat and protein (19% fat and 34% protein among the males and 15% fat and 32% protein among the females). Males consumed a

significantly higher percentage of energy from protein than the females (P value <0.05). Females consumed a higher percentage of energy from carbohydrates than males (53% and 47%, respectively). With macronutrient intake guidelines, more females than males were within the acceptable macronutrient distribution range (AMDR). All participants were within the AMDR for protein and above the EAR for carbohydrates and protein. Significant differences were seen between male and female participants that were within the AMDR for carbohydrates (P<0.01).

For thiamin, riboflavin, niacin, vitamin B<sub>6</sub>, and selenium, no participants were below the EAR for both genders (**Table 4.6**). There were only female participants that are below the guideline for vitamin B<sub>12</sub> and folate. Females had significantly higher percentages for intakes below the guideline for vitamins D and E (P <0.05). Nearly all female participants (96%) were consuming intakes below the recommended amount for vitamin D and 84% were below EAR for vitamin E. Intake of vitamin A is the only nutrient having more males than females consuming inadequate amounts; however, the difference is not significant. Overall, females in this population are consuming more micronutrients under the recommended intake than males.

#### *4.3.2. Supplement types and use*

Overall, 39% of the population in this study uses one or more dietary supplements. A larger percentage of the male population was classified as supplement users (53% males and 30% females). Data on the types of supplements consumed in **Figure 4.4** demonstrates that multivitamins were most

commonly consumed among females while protein or individual amino acids were more common with male participants. Significantly more males consumed protein supplement than females ( $P$  value $<0.05$ ). Supplements only consumed by male users included soy protein, melatonin, quercetin, fish oils, herbal complex, green tea supplement, and caffeine anhydrous. Supplements found only among female users were fiber and calcium. **Table 4.7** compares dietary intake between supplement users and supplement non-users from diet only as well as comparing diet only among users to dietary + supplement intake. In addition, nutrient adequacy was assessed among non-users, users from diet only, and users from diet + supplement intake. Supplement users had significantly higher intakes from diet only for protein, folate, niacin, vitamin E, magnesium, and zinc intakes than non-users ( $P<0.05$ ).

With the addition of a supplement among users, all nutrients except vitamin A were significantly higher than non-users ( $P<0.05$ ). For vitamin D, vitamin C, and zinc intakes, the addition of a supplement to dietary intakes among users significantly increased average intakes ( $P>0.05$ ). For all nutrients included, intakes from the diet for supplement users were greater than non-users and, therefore, had fewer individuals below the guideline (except vitamin C). For protein, folate, niacin, and zinc intakes, supplement users had no individuals under the EAR for average dietary intakes. For protein, niacin, and vitamin B<sub>12</sub>, non-users had no individuals under the EAR for average dietary intakes. Percentages of non-users below the EAR for dietary intakes of vitamin D, vitamin E, and magnesium were significantly higher than users compared to dietary

intake only and with the addition of a supplement ( $P<0.05$ ). When comparing adequacy from dietary sources only and total intake including supplements among users only, supplementation significantly decreased the percentage of individuals below the guideline for only vitamin D. For most nutrients, excluding folate, niacin, iron, and zinc, average consumption from non-users and users was below the upper limit (UL). Supplement users had more individuals consuming above the UL than non-users for folate, niacin, and iron ( $P<0.05$ ). For niacin, supplement intake increased the percentage of individuals above the UL significantly when compared to percentage above for diet only ( $P<0.05$ ).

**Table 4.1.** Characteristics and misreporting of 60 male and female college students from UCONN

	Males (n=20)	Females (n=40)
<i>Age, years</i> <sup>1</sup>	20.4±2.2	19.5±1.4
<i>BMI</i> <sup>1,2</sup>	23.9±3.1	22.7±2.4
<i>Energy, kcal/d</i> <sup>1</sup>	2,526±711	1,834±498
<i>BMR</i> <sup>1,3</sup>	1,814±141	1,381±130
<i>EI:BMR<sub>est</sub></i> <sup>1,4</sup>	1.4±0.4	1.3±0.4
<i>EI:BMR<sub>est</sub>, %</i>		
<1.12, underreporting	15	30
>2.28, overreporting	0	2.5

<sup>1</sup> Values are presented as mean ± SD

<sup>2</sup> BMI, Body Mass Index kg/m<sup>2</sup>

<sup>3</sup> Basal Metabolic Rate. Schofield equations for sex and age (18-30)  
Males: BMR=15.0x weight(kg) + 690. Females: BMR=14.8x weight(kg) + 485.

<sup>4</sup> Energy Intake: Basal Metabolic Rate estimated ratio



**Table 4.2.** Lifestyle characteristics of 44 male and female college students from UCONN

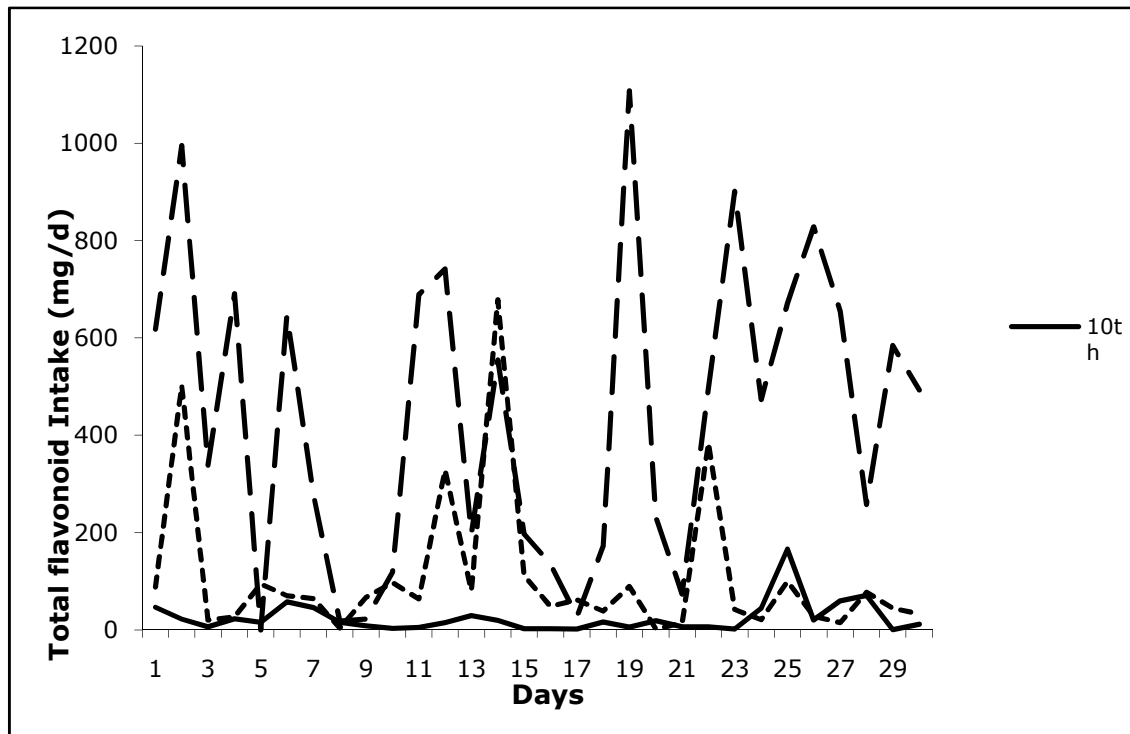
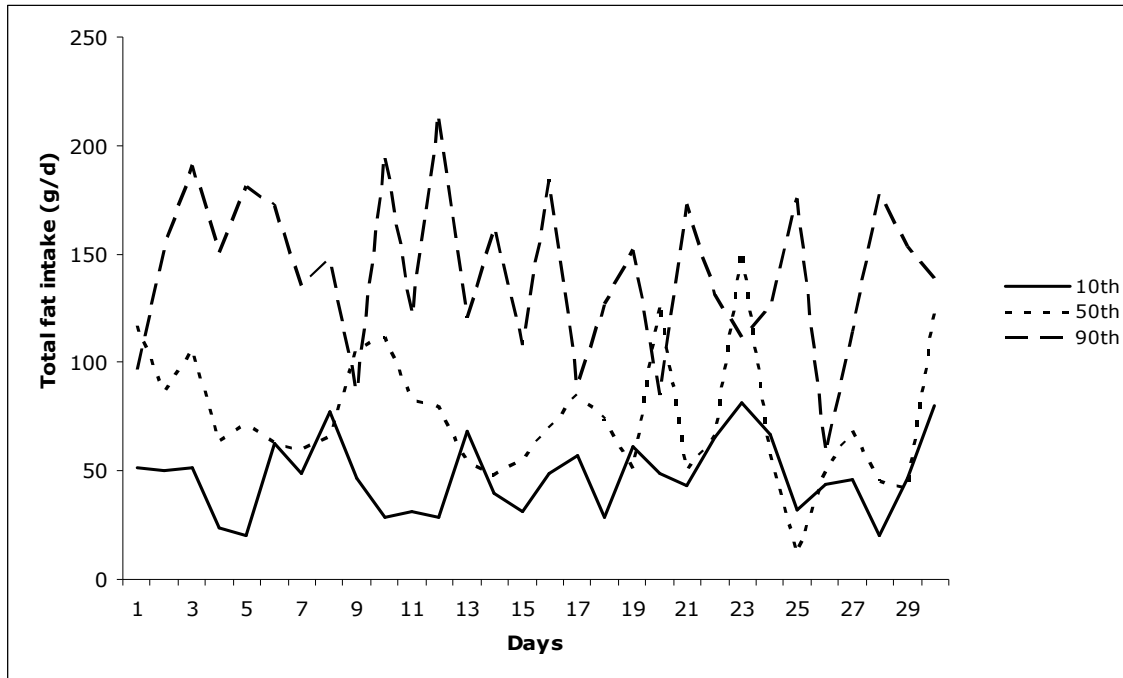
	Males (n=17)	Females (n=27)
<i>Age, years<sup>1</sup></i>	20.4±2.2	19.6±1.5
<i>BMI<sup>2</sup></i>	24.1±2.9	22.2±2.6
<i>Ethnicity, %<sup>3</sup></i>		
Non Hispanic White	82	74
Non Hispanic Black	6	4
Hispanic	0	11
Asian or Island Pacificer	12	11
<i>Year, %<sup>3</sup></i>		
Freshman	24	33
Sophomore	35	30
Junior	6	11
Senior	18	26
Graduate	18	0
<i>Moderate Activity &gt;30 minutes/5 days, %<sup>3,4</sup></i>	59	81
<i>Vigorous Activity &gt;20 minutes/ 3 days, %<sup>3,4</sup></i>	88	70
<i>Health Science Major, %<sup>3</sup></i>	18	48
<i>Consume Campus Meals, %<sup>3</sup></i>	64	74
<i>Consume Fast Food, %<sup>3</sup></i>	24	37
<i>Pressure to be a certain weight, %<sup>3</sup></i>	18	63
<i>Fear of gaining weight, %<sup>3</sup></i>	18	37
<i>Skip meals to lose weight, %<sup>3</sup></i>	0	30

<sup>1</sup> Values are presented as mean ± SD

<sup>2</sup> BMI, Body Mass Index kg/m<sup>2</sup>

<sup>3</sup> Percents for were answer yes to questions in Health and Nutrition Survey

<sup>4</sup> Exercise guidelines established by ACSM and AHA for average healthy adult



**Figure 4.1.** Daily intakes for three college students from UCONN at the 10<sup>th</sup>, 50<sup>th</sup>, and 90<sup>th</sup> percentiles of distribution for total fat intake (A) and total flavonoid intakes (B).

**Table 4.3.** Means, coefficients of variation, and variance ratios for energy and nutrient intakes among 44 college students at UCONN

Nutrient	Mean±SD	CV <sub>w</sub> <sup>a</sup>	CV <sub>b</sub> <sup>b</sup>	Variance Ratio <sup>c</sup>
<i>Energy, kcal/d</i>	2,274±583	30.2	20.5	2.18
<i>Total Fat, g/d</i>	84±30	47.2 (27.4)	30.6 (18.8)	2.38 (2.12)
<i>Total Carbohydrate, g/d</i>	284±71	29.4 (18.4)	23.1 (16.0)	1.62 (1.32)
<i>Protein, g/d</i>	95±43	34.1 (25.4)	35.8 (24.3)	0.91 (1.10)
<i>Vitamin A, µg RAE/d</i>	992.4±469.1	59.1 (59.1)	43.4 (43.4)	1.86 (1.86)
<i>β-carotene, µg/d</i>	4,396.4±3,059.2	119.2 (119.2)	66.8 (66.80)	3.18 (3.18)
<i>α-carotene, µg/d</i>	726.5±625.6	188.4 (188.5)	81.2 (81.0)	5.39 (5.41)
<i>β-cryptoxanthin, µg/d</i>	218.6±196.4	278.3 (278.3)	85.6 (85.9)	10.56 (10.51)
<i>Lutein + Zeaxanthin, µg/d</i>	2,727.9±1,925.0	163.9 (163.8)	74.3 (74.0)	4.87 (4.91)
<i>Lycopene, µg/d</i>	6,656.9±6,134.7	124.0 (123.0)	99.6 (100.0)	1.55 (1.51)
<i>Carotenoids, µg/d</i>	14,862.9±9,464.4	82.8 (82.5)	60.7 (60.6)	1.86 (1.85)
<i>Thiamin, mg/d</i>	2.1±0.7	42.2 (41.6)	27.8 (27.4)	2.30 (2.31)
<i>Riboflavin, mg/d</i>	2.7±1.3	39.3 (39.5)	43.1 (43.3)	0.83 (0.83)
<i>Niacin, mg/d</i>	30.1±15.4	45.1 (45.1)	42.9 (42.9)	1.10 (1.11)
<i>Vitamin B<sub>6</sub>, mg/d</i>	2.9±1.3	62.7 (61.7)	74.0 (72.7)	0.72 (0.71)
<i>Folate, µg DFE/d</i>	613.9±49.1	53.5 (53.5)	48.9 (48.9)	1.19 (1.19)
<i>Vitamin B<sub>12</sub>, µg/d</i>	6.9±5.4	128.5 (129.2)	62.4 (62.7)	4.23 (4.23)
<i>Vitamin C, mg/d</i>	125.3±95.4	64.8 (64.4)	39.6 (38.7)	2.68 (2.76)
<i>Vitamin D, µg/d</i>	5.3±3.9	78.9 (78.9)	64.5 (64.5)	1.50 (1.50)
<i>α-Tocopherol, mg/d</i>	14.7±11.0	85.0 (81.0)	65.6 (59.9)	1.68 (1.83)
<i>γ-Tocopherol, mg/d</i>	13.5±5.2	82.6 (68.9)	35.0 (31.0)	5.57 (4.94)
<i>Total Vitamin E, mg/d</i>	31.4±13.3	65.0 (53.1)	35.1 (26.8)	3.43 (3.93)
<i>Calcium, mg/d</i>	1,074.9±438.7	41.1 (41.1)	37.4 (37.4)	1.21 (1.21)
<i>Iron, mg/d</i>	20.9±10.6	54.2 (54.8)	42.8 (43.3)	1.60 (1.61)
<i>Magnesium, mg/d</i>	355.9±156.1	35.3 (35.3)	37.1 (37.1)	0.91 (0.91)
<i>Selenium, mg/d</i>	130.4±49.1	40.4 (40.4)	28.1 (28.1)	2.07 (2.07)
<i>Zinc, mg/d</i>	12.8±5.5	48.7 (48.8)	34.1 (34.1)	2.04 (2.04)
<i>Total Flavonoids, mg/d</i>	165.7±201.7	154.4 (154.3)	118.2 (117.5)	1.70 (1.73)
<i>Isoflavones, mg/d</i>	4.4±5.3	258.5 (258.2)	110.3 (111.6)	5.49 (5.35)
<i>Proanthocyanidins, mg/d</i>	110.0±114.7	130.4 (129.7)	100.4 (103.2)	1.69 (1.58)

Note: Values in parentheses are adjusted for energy intake

<sup>a</sup> $[(\sqrt{\text{within-person variation}})/\text{mean}] \times 100$

<sup>b</sup> $[(\sqrt{\text{between-person variation}})/\text{mean}] \times 100$

<sup>c</sup> $(\text{within-person variation}/\text{between-person variation}) = (\text{CV}_w/\text{CV}_b)^2$

**Table 4.4.** Means, coefficients of variation, and variance ratios for energy and nutrient intakes among 44 male and female college students at UCONN

Nutrient	Mean±SD	CV <sub>w</sub> <sup>a</sup>	CV <sub>b</sub> <sup>b</sup>	Variance Ratio <sup>c</sup>
<i>Energy, kcal/d</i>				
Male	2,695±622	31	22.4	1.92
Female	2,009±367*	28.5	17.3	2.7
<i>Total Fat, g/d</i>				
Male	103±61	49.8 (27.5)	31.7 (20.6)	2.47 (1.78)
Female	72±20*	41.7 (25.6)	26.0 (16.0)	2.58 (2.57)
<i>Total Carbohydrate, g/d</i>				
Male	315±85	31.1 (19.7)	25.3 (19.9)	1.51 (0.97)
Female	264±54*	27.6 (17.0)	19.4 (11.1)	2.01 (2.36)
<i>Protein, g/d</i>				
Male	127±52	32.9 (24.0)	39.1 (26.6)	0.71 (0.82)
Female	74±16*†	32.7 (25.1)	20.0 (13.2)	2.67 (3.61)
<i>Vitamin A, µg RAE/d</i>				
Male	1,112.5±578.8	60.9 (59.4)	50.8 (45.9)	1.44 (1.67)
Female	884.6±355.9*	59.0 (57.7)	38.8 (36.8)	2.31 (2.46)
<i>β-carotene, µg/d</i>				
Male	4,665.5±3,303.5	119.4 (119.5)	65.1 (67.5)	3.36 (3.14)
Female	4,226.9±2,947.1	118.7 (118.5)	64.9 (66.4)	3.35 (3.19)
<i>α-carotene, µg/d</i>				
Male	745.9±712.9	193.9 (194.0)	85.7 (89.1)	5.11 (4.74)
Female	714.3±591.6	184.5 (184.5)	74.0 (74.3)	6.22 (6.16)
<i>β-cryptoxanthin, µg/d</i>				
Male	226.4±289.5	358.1 (358.5)	105.4 (109.2)	11.54 (10.77)
Female	213.7±158.0	203.5 (203.4)	62.3 (63.8)	10.66 (10.17)
<i>Lutein + Zeaxanthin, µg/d</i>				
Male	2,830.6±2,025.3	190.4 (190.5)	60.1 (61.4)	10.03 (9.63)
Female	2,663.4±2,277.6	142.0 (141.9)	79.8 (81.7)	3.16 (3.01)
<i>Lycopene, µg/d</i>				
Male	8,874.4±10,352.0	103.3 (102.8)	111.6 (115.4)	0.86 (0.79)
Female	5,260.8±2,955.2	144.8 (143.2)	48.4 (50.5)	8.96 (8.04)
<i>Carotenoids, µg/d</i>				
Male	17,700.3±12,626.3	77.0 (77.0)	67.8 (69.9)	1.29 (1.21)
Female	13,076.4±6,438.8	87.1 (85.9)	45.6 (46.9)	3.65 (3.35)
<i>Thiamin, mg/d</i>				
Male	2.5±0.8	46.7 (36.1)	31.1 (20.0)	2.25 (3.25)
Female	1.8±0.5*	34.7 (31.6)	24.3 (19.0)	2.03 (2.77)
<i>Riboflavin, mg/d</i>				
Male	3.4±1.7	40.9 (34.5)	49.7 (37.1)	0.68 (0.87)
Female	2.3±0.8*†	34.6 (31.9)	32.4 (28.4)	1.14 (1.26)
<i>Niacin, mg/d</i>				
Male	39.9±20.2	45.3 (39.7)	49.8 (39.3)	0.83 (1.02)

Female	23.9±6.6* <sup>†</sup>	40.7 (37.6)	26.7 (21.8)	2.33 (2.97)
<i>Vitamin B<sub>6</sub>, mg/d</i>				
Male	4.1±3.4	62.1 (60.3)	83.3 (77.8)	0.55 (0.60)
Female	2.1±0.6* <sup>†</sup>	50.8 (49.1)	28.4 (22.9)	3.21 (4.62)
<i>Folate, µg DFE/d</i>				
Male	811.1±483.8	56.6 (53.7)	58.7 (53.8)	0.93 (0.99)
Female	489.8±119.6* <sup>†</sup>	42.3 (40.7)	23.2 (19.5)	3.33 (4.37)
<i>Vitamin B<sub>12</sub>, µg/d</i>				
Male	10.2±6.8	122.3 (121.5)	64.5 (55.8)	3.59 (4.74)
Female	5.3±3.1* <sup>†</sup>	117.2 (116.8)	48.3 (44.7)	5.89 (6.82)
<i>Vitamin C, mg/d</i>				
Male	144.9±60.1	66.3 (66.2)	39.9 (40.6)	2.76 (2.66)
Female	122.1±46.4	62.6 (61.8)	36.9 (36.7)	2.88 (2.83)
<i>Vitamin D, µg/d</i>				
Male	7.4±4.8	77.4 (75.0)	63.9 (53.1)	1.46 (1.99)
Female	4.0±3.9* <sup>†</sup>	2.3 (2.2)	2.0 (1.9)	1.29 (1.39)
<i>α-Tocopherol, mg/d</i>				
Male	21.3±15.3	86.0 (82.0)	68.1 (63.3)	1.60 (1.68)
Female	10.2±3.1* <sup>†</sup>	58.7 (54.8)	27.6 (24.9)	4.51 (4.85)
<i>γ-Tocopherol, mg/d</i>				
Male	14.9±4.9	90.6 (70.1)	27.1 (35.3)	11.13 (3.94)
Female	12.6±5.3	74.3 (66.7)	38.9 (30.4)	3.64 (4.83)
<i>Total Vitamin E, mg/d</i>				
Male	39.7±15.6	67.8 (53.8)	36.2 (29.1)	3.52 (3.41)
Female	26.1±8.3*	58.5 (49.6)	30.0 (22.9)	3.82 (4.69)
<i>Calcium, mg/d</i>				
Male	1,254.2±551.4	42.5 (37.8)	43.3 (32.8)	0.97 (1.32)
Female	962.1±311.4*	38.8 (34.9)	31.6 (26.5)	1.51 (1.73)
<i>Iron, mg/d</i>				
Male	27.3±14.4	57.4 (52.5)	51.8 (42.1)	1.23 (1.56)
Female	16.9±3.9* <sup>†</sup>	44.0 (41.7)	21.6 (18.4)	4.16 (5.15)
<i>Magnesium, mg/d</i>				
Male	452.2±193.4	37.2 (29.3)	42.2 (33.8)	0.78 (0.75)
Female	295.3±85.9* <sup>†</sup>	30.1 (25.6)	28.5 (26.2)	1.11 (0.95)
<i>Selenium, mg/d</i>				
Male	201.3±77.3	34.2 (26.6)	27.0 (14.1)	1.61 (3.55)
Female	106.2±44.6* <sup>†</sup>	37.0 (30.1)	20.2 (13.4)	3.34 (5.02)
<i>Zinc, mg/d</i>				
Male	16.6±15.9	50.0 (44.9)	34.1 (25.4)	2.15 (3.13)
Female	10.4±3.7* <sup>†</sup>	43.3 (40.7)	34.2 (29.5)	1.60 (1.90)
<i>Total Flavonoids, mg/d</i>				
Male	236.9±250.3	145.0 (145.0)	102.2 (103.8)	2.01 (1.94)
Female	120.9±152.7 <sup>†</sup>	148.3 (148.4)	124.4 (127.0)	1.42 (1.36)
<i>Isoflavones, mg/d</i>				
Male	5.6±6.3	221.3 (220.9)	101.4 (105.7)	4.77 (4.37)
Female	3.7±4.5	288.7 (288.7)	108.2 (112.1)	7.12 (6.63)

*Proanthocyanidins,*  
*mg/d*

Male	139.8±91.3	125.8 (125.6)	113.2 (119.2)	1.24 (1.11)
Female	91.3±61.4	130.0 (128.4)	61.5 (63.6)	4.5 (4.1)

Note: Values in parentheses are adjusted for energy intake

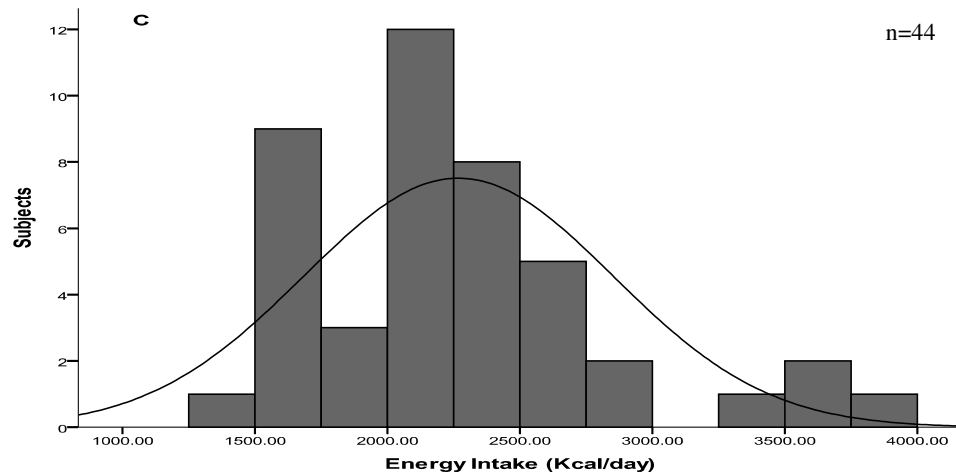
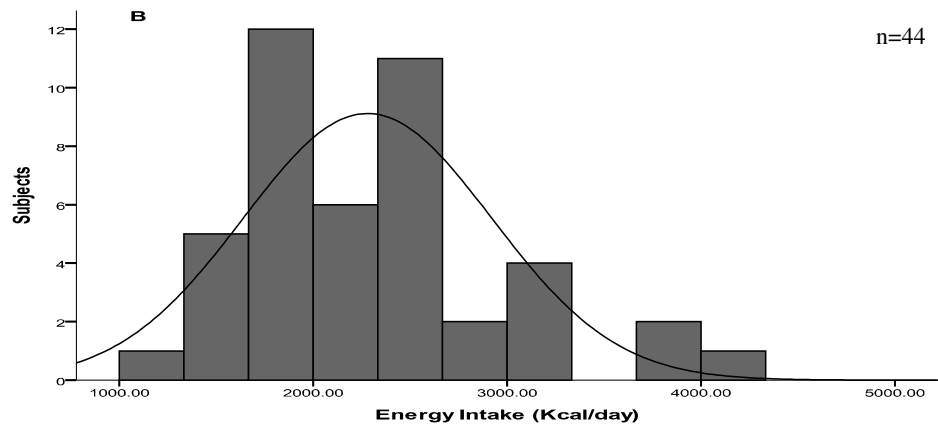
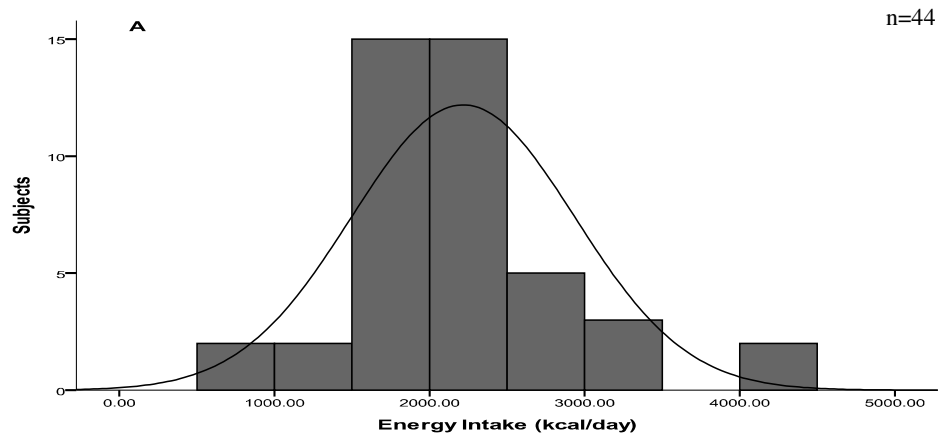
<sup>a</sup> $[(\sqrt{\text{within person variation}})/\text{mean}] \times 100$

<sup>b</sup> $[(\sqrt{\text{between person variation}})/\text{mean}] \times 100$

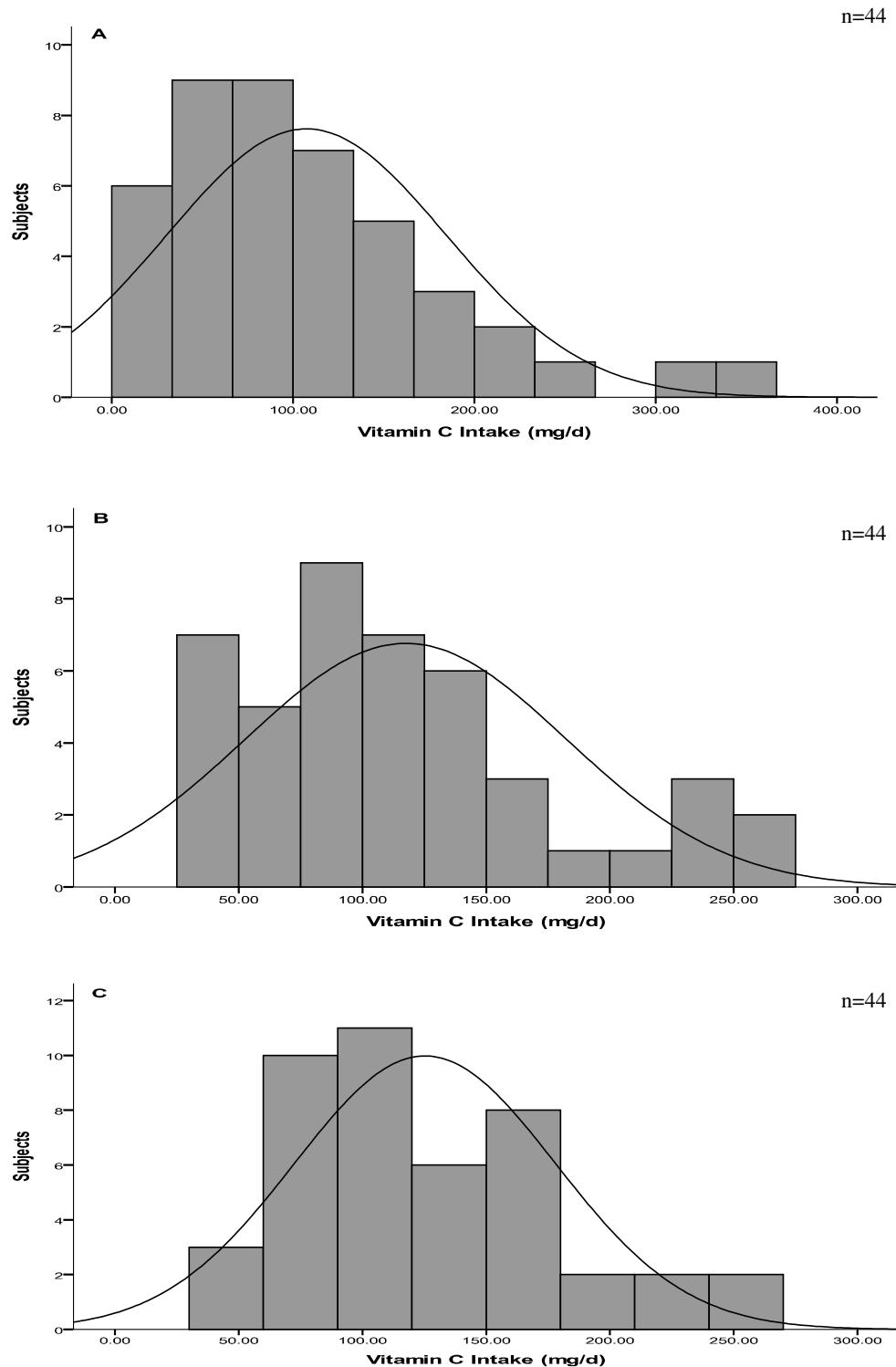
<sup>c</sup> $(\text{within-person variation}/\text{between-person variation}) = (CV_w/CV_b)^2$

\* Different from males, P value <0.05

† Different from males after adjusted for energy intake, P value <0.05

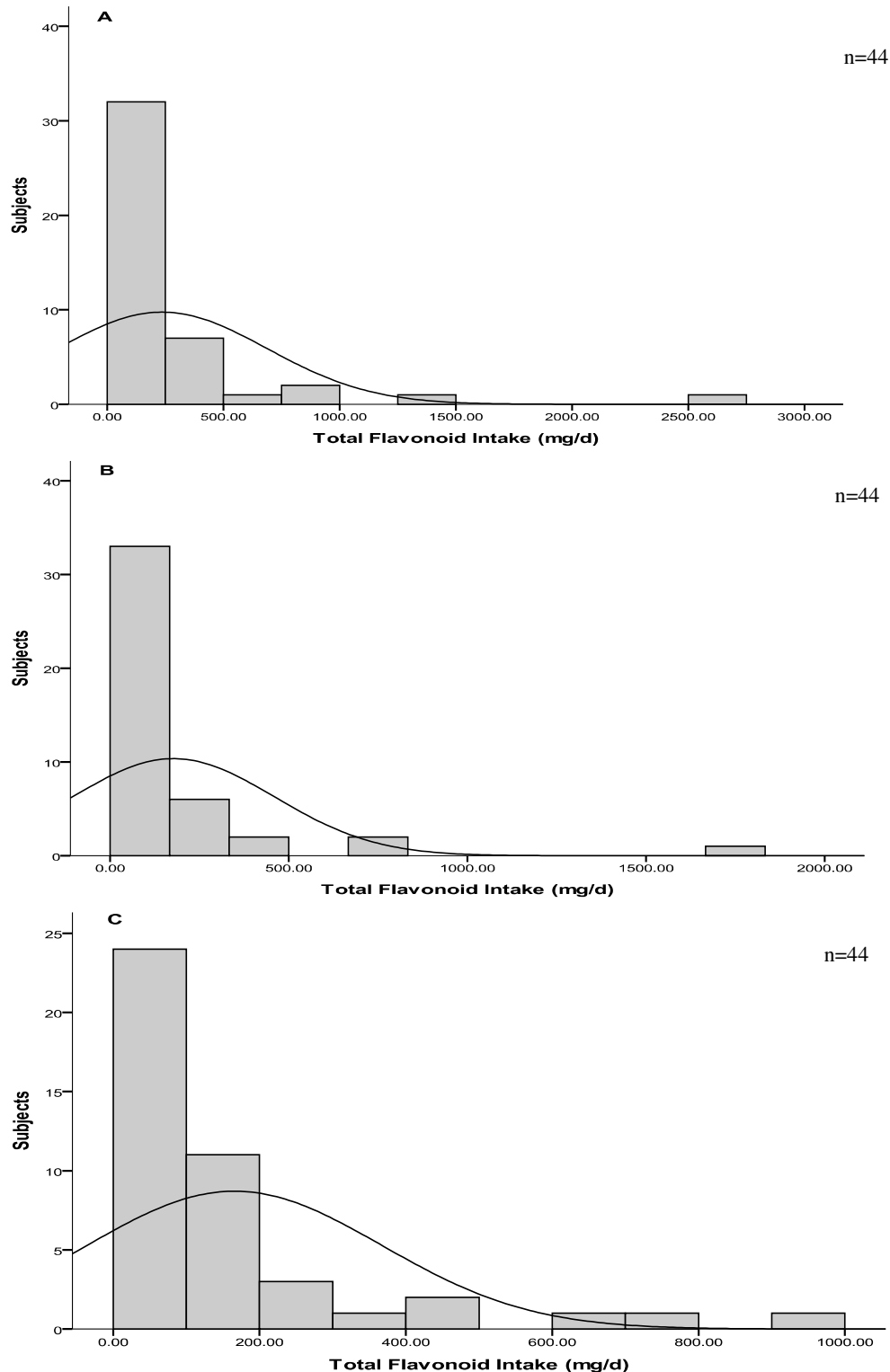


**Figure 4.2-1.** Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of energy intake for 44 college students from UCONN. Mean energy intake ( $\pm$ S.D.) for 1 day was  $2,216 \pm 720$  kcal/d,  $2,280 \pm 642$  kcal/d for 7 days, and  $2,274 \pm 583$  kcal/d for 30 days, respectively.

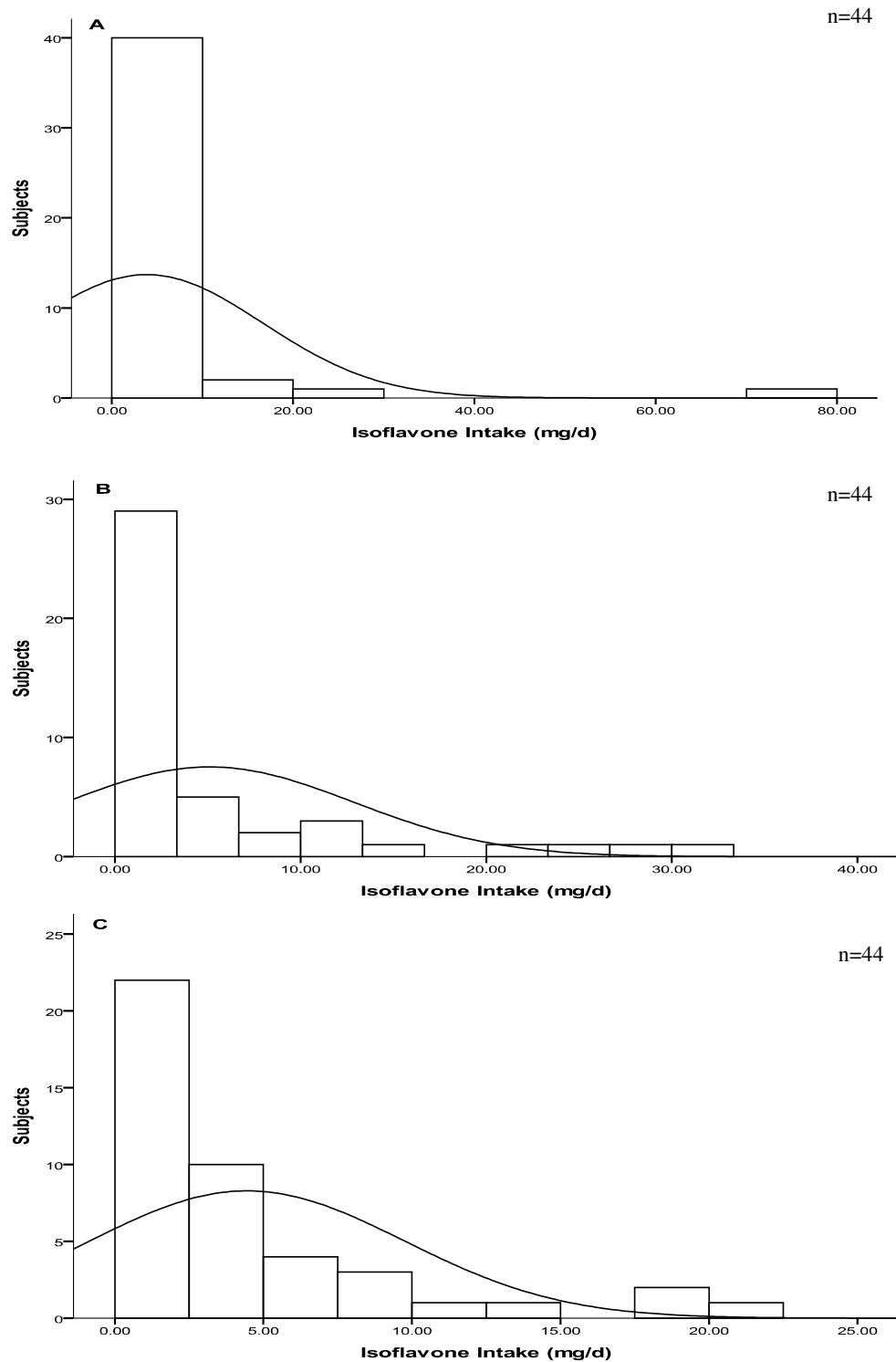


**Figure 4.2-2.** Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of vitamin C intake for 44 college students from UCONN. Mean vitamin C intake ( $\pm$ S.D.) for 1 day was  $107.3 \pm 76.8$  mg/d,  $117.6 \pm 64.9$  mg/d for 7 days, and  $130.9 \pm 52.7$  mg/d for 30 days, respectively.

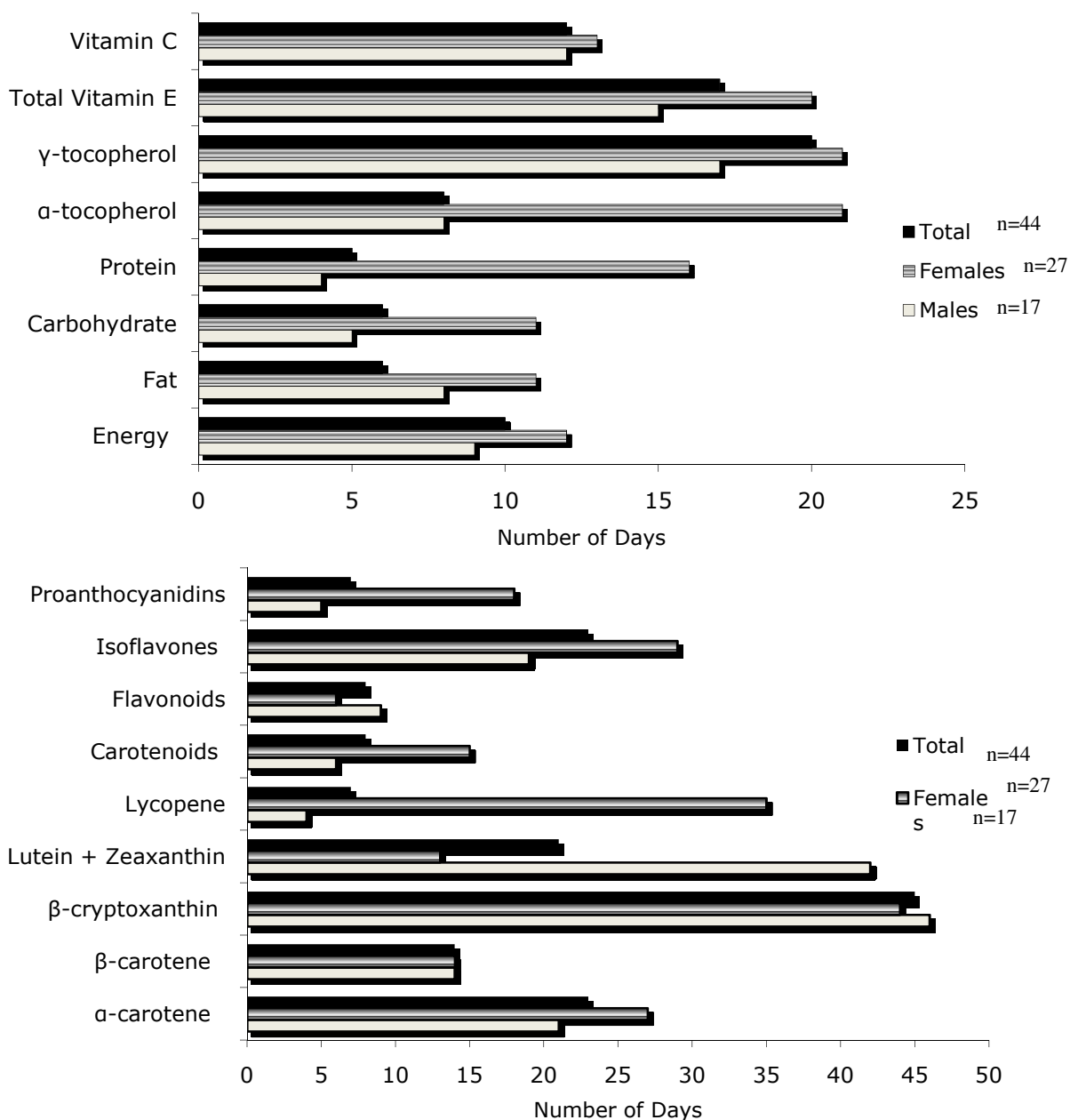




**Figure 4.2-3.** Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of total flavonoid intake for 44 college students from UCONN. Mean total flavonoid intake ( $\pm$ S.D.) for 1 day was 236.4 $\pm$ 449.9 mg/d, 177.2 $\pm$ 282.5 mg/d for 7 days, and 165.7 $\pm$ 201.7 mg/d for 30 days, respectively.



**Figure 4.2-4.** Effects of randomly selecting 1 day (A), 7 days (B), and 30 days (C) per subject on the mean distribution of isoflavone intake for 44 college students from UCONN. Mean isoflavone intake ( $\pm$ S.D.) for 1 day was  $3.9 \pm 12.8$  mg/d,  $5.1 \pm 7.8$  mg/d for 7 days, and  $4.5 \pm 5.3$  mg/d for 30 days, respectively.



**Figure 4.3.** The number of days of dietary assessment required to estimate usual macronutrient and antioxidant intakes among total, male, and female college students from UCONN after adjusting for energy intake and gender for total population and energy intake among genders, respectively

**Table 4.5.** The number of days of dietary assessment required to estimate usual micronutrient intakes among 44 male and female college students from UCONN

Nutrient	Days <sup>1,2</sup>	
	Male	Female
<i>Vitamin A, µg RAE/d</i>	8	11
<i>Thiamin, mg/d</i>	14	12
<i>Riboflavin, mg/d</i>	4	6
<i>Niacin, mg/d</i>	5	13
<i>Vitamin B<sub>6</sub>, mg/d</i>	3	20
<i>Folate, µg DFE/d</i>	5	19
<i>Vitamin B<sub>12</sub>, µg/d</i>	21	25
<i>Vitamin C, mg/d</i>	12	13
<i>Vitamin D, µg/d</i>	9	6
<i>α-Tocopherol, mg/d</i>	8	21
<i>Calcium, mg/d</i>	6	8
<i>Iron, mg/d</i>	7	22
<i>Magnesium, mg/d</i>	4	5
<i>Selenium, mg/d</i>	16	22
<i>Zinc, mg/d</i>	14	9

<sup>1</sup> All values are adjusted for energy intake

<sup>2</sup> number of days calculated from  $= [r^2 / (1 - r^2)] \times (S_w^2 / S_b^2)$

**Table 4.6.** Nutrient intake adequacy for select nutrients among 44 male and female college students from UCONN

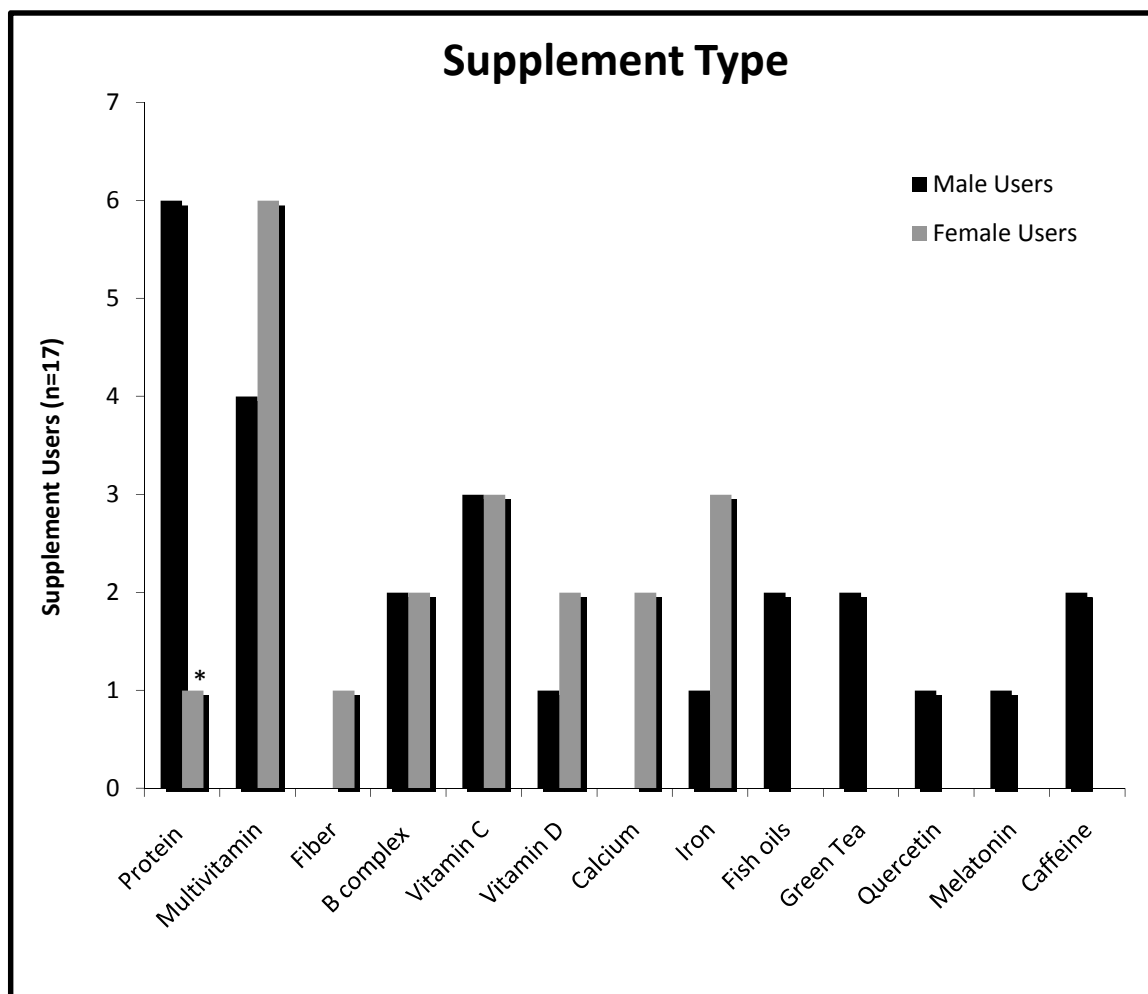
Nutrient <sup>1</sup>	Male (n=17)		Female (n=27)	
		% within AMDR <sup>2,3</sup>		% within AMDR <sup>2,3</sup>
Energy, kcal/d	2,695±622	-	2,009±367	-
Fat, % of energy	34	59	32	78
Carbohydrates, % of energy	47	53	53	93
Protein, % of energy	19	100	15*	100
	% below EAR <sup>2,3</sup>		% below EAR <sup>2,3</sup>	
Carbohydrates, g/d	315±85	0	264±54	0
Protein, g/d	127±52	0	74±16*	0
Vitamin A, µg RAE/d	1,112.5±578.8	24	884.6±355.9	8
Thiamin, mg/d	2.5±0.8	0	1.8±0.5	0
Riboflavin, mg/d	3.4±1.7	0	2.3±0.8*	0
Niacin, mg/d	39.9±20.2	0	23.9±6.6*	0
Vitamin B <sub>6</sub> , mg/d	4.1±3.4	0	2.1±0.6*	0
Folate, µg DFE/d	811.1±483.8	0	489.8±119.6*	4
Vitamin B <sub>12</sub> , µg/d	10.2±6.8	0	5.3±3.1*	4
Vitamin C, mg/d	144.9±60.1	12	122.1±46.4	4
Vitamin D, µg/d	7.4±4.8	76	4.0±3.9*	96*
Vitamin E, mg/d	21.3±15.3	41	10.2±3.1*	84*
Calcium, mg/d	1,254.2±551.4	29	962.1±311.4	41
Iron, mg/d	27.3±14.4	-	16.9±3.9*	-
Magnesium, mg/d	452.2±193.4	29	295.3±85.9*	41
Selenium, mg/d	201.3±77.3	0	106.2±44.6*	0
Zinc, mg/d	16.6±15.9	12	10.4±3.7*	4

<sup>1</sup> Values presented as untransformed mean ± SD

<sup>2</sup> Dietary Reference Intakes (DRIs) for select nutrients by life stage group established by the Institute of Medicine (IOM) (59,112).

<sup>3</sup> Percents below EAR are based on transformed mean values for each nutrient based on EAR cut-point method (117).

\* Significantly different from males after energy adjustment, P value <0.05



**Figure 4.4.** Form of supplement consumed by 17 male and female supplement users from UCONN. \*Significant difference from male users according to chi-squared analysis, P value <0.05

**Table 4.7.** Comparison of nutrient intake adequacy between dietary supplement users and non users from UCONN.

Nutrient <sup>1,2</sup>	Non-users		Users
	n=27	diet only n=17	dietary + supplement intake n=17
<i>Protein, g/d</i>	83.6±28.9	112.0±55.3 <sup>4</sup>	115.0±56.9 <sup>4</sup>
% below EAR <sup>3</sup>	0	0	0
<i>Vitamin A, µg RAE/d</i>	940.2±63.9	1,024.2±469.4	1,211.6±518.6
% below EAR <sup>3</sup>	11	24	24
<i>Folate, µg DFE/d</i>	505.6±184.8	750.1±494.9 <sup>4</sup>	851.4±465.9 <sup>4</sup>
% below EAR <sup>3</sup>	4	0	0
% below UL <sup>3</sup>	4	18	24 <sup>4</sup>
<i>Niacin, mg/d</i>	25.4±7.5	37.4±21.5 <sup>4</sup>	40.9±20.6 <sup>4</sup>
% below EAR <sup>3</sup>	0	0	0
% below UL <sup>3</sup>	15	41 <sup>4</sup>	59 <sup>4,5</sup>
<i>Vitamin B<sub>12</sub>, µg/d</i>	6.3±4.0	8.6±6.9	16.1±20.6 <sup>4</sup>
% below EAR <sup>3</sup>	7	6	6
<i>Vitamin C, mg/d</i>	127.1±48.7	137.0±59.4	180.5±72.2 <sup>4,5</sup>
% below EAR <sup>3</sup>	4	12	6
<i>Vitamin D, µg/d</i>	4.6±3.3	6.4±4.6	13.5±9.9 <sup>4,5</sup>
% below EAR <sup>3</sup>	96	76 <sup>4</sup>	47 <sup>4,5</sup>
<i>Vitamin E, mg α-tocopherol/d</i>	11.1±5.0	20.4±15.1 <sup>4</sup>	27.6±17.8 <sup>4</sup>
% below EAR <sup>3</sup>	81	41 <sup>4</sup>	18 <sup>4</sup>
<i>Calcium, mg/d</i>	938.3±328.5	1,220.6±552.4	1,364.4±567.7 <sup>4</sup>
% below EAR <sup>3</sup>	37	29	29
<i>Magnesium, mg/d</i>	411.3±631.7	450.7±186.8 <sup>4</sup>	463.4±183.7 <sup>4</sup>
% below EAR <sup>3</sup>	56	12 <sup>4</sup>	12 <sup>4</sup>
<i>Iron, mg/d</i>	18.4±4.7	24.9±15.5	32.6±22.6 <sup>4</sup>
% below UL <sup>3</sup>	0	18 <sup>4</sup>	18 <sup>4</sup>
<i>Zinc, mg/d</i>	11.1±3.7	15.6±6.8 <sup>4</sup>	20.4±6.6 <sup>4,5</sup>
% below EAR <sup>3</sup>	11	0	0
% below UL <sup>3</sup>	0	0	6
<i>Total flavonoids, mg/d</i>	128.3±158.6	213.7±252.6	286.9±352.6 <sup>4</sup>

<sup>1</sup> Values are presented as means ± SD.

<sup>2</sup> Dietary Reference Intakes (DRIs) for select nutrients by life stage group established by the Institute of Medicine (IOM) (59, 112).

<sup>3</sup> Percents below EAR are based on transformed mean values for each nutrient based on EAR cut-point method (117).

<sup>4</sup> Significantly different from non users after energy and gender adjustment, P value <0.05

<sup>5</sup> Significantly different among users only after energy and gender adjustment, P value <0.05

## **Chapter 5**

### **Discussion**

#### **5.1. Identification of Misreporting and Estimation of Usual Intakes for Energy and Key Nutrients, Including Antioxidants, among College Students**

The major findings for this objective were that more females than males were classified as under- and over-reporters as well as the within person variation among the females was greater than males for most nutrients.

##### *5.1.1. Misreporting among Male and Female College Students*

Despite the well documented importance of defining participants in a study who misreport intakes to the accuracy of the data (40, 42, 96), few studies employ these procedures before reporting population intakes. A recent review of major determinants of misreporting energy intakes in nutritional studies reports that BMI, age, gender, socioeconomic status and education, smoking and dieting practices, psychological factors such as depression, and eating habits are the most common predictors of underreporting (94). The review reports that more females than males underreport and this behavior increases as BMI and age increases. The present study had similar results to a study with non-obese, weight stable adults with 14% of males and 49% of females classified as underreporters (89). Whether males tend to underreport less than women or if higher energy requirements of men allow them to rarely fall below the cutoff limits when applied to an entire study population is not known (93). However, it is thought that underreporting in females may be attributed to psychological issues



with weight and body perception (161). Issues with body dissatisfaction, low self esteem, and fat avoidance behaviors are prevalent among women (95). From the questions relating to body image in the health and nutrition survey, 58% of the females who underreported nutrient intake selected that they felt pressured to be a certain weight and 67% indicate that their weight moderately to extremely affected their view of self. These predictors among the females are consistent with those noted in a review by Maurer et al. despite the differences in survey methods (162). These body weight related issues were not present in the male underreporters. Among the underreporters, 75% of the females had a BMI of 23 to 25. While this is classified as a healthy body weight, only 33% of the remaining females who did not under report energy intake had a BMI over 23. Therefore, in this population, the major predictors of underreporting were gender (being female), psychological issues related to body image, and BMI. The majority of misreporters were classified as underreporters, 2.5% of the females were identified as overreporters. However, due to the seldom occurrence of overreporting in populations compared to underreporting, less emphasis is given to addressing this issue in the literature (94). Future research should address the issue of any bias towards identifying mainly underreporters in nutrient intake data. Overall, despite the heavy burden of long term dietary assessment, there was a low prevalence of misreporting among this college population.

### *5.1.2. Mean Intakes of Energy and Key Nutrients among Male and Female College Students*

The males in the present study were consuming higher intakes for all nutrients except for several antioxidant nutrients (carotenoids, vitamin C,  $\gamma$ -tocopherol, and the polyphenols). After adjusting for energy intake, the consumption of protein, the B vitamins (excluding thiamin), vitamin D,  $\alpha$ -tocopherol, magnesium, iron, selenium, zinc, and flavonoids were higher in males than females. The findings of major interest were the differences between gender intakes for  $\alpha$ -tocopherol and flavonoids. Further analysis of the vitamin E intakes indicate that the intakes for  $\gamma$ -tocopherol were similar between the genders. We noted that many of the males were consuming large quantities of fortified cereals which are rich sources of  $\alpha$ -tocopherol. Many grains are fortified in the U.S. with  $\alpha$ -tocopherol due to the fact that it is the only form of vitamin E that has a DRI (59). The main sources of flavonoids among the males were tea, wine, and vegetables such as lettuce, onions and peppers, and fruits such as apples, citrus fruits and juices. The main sources among the females were similar with tea, wine, mixed salads and lettuce, and fruits such as citrus fruits and juices, bananas, melons, and grapes being the top contributors. These dietary sources are consistent with previous data from our laboratory and those from analysis of NHANES data (80, 82, 104). The major difference between the genders was the quantity of tea consumed among the males which is in contrast to data from NHANES 1999-2002 that reported tea consumption was higher among older females (163).

In comparison to usual nutrient intakes among other young adult populations, the macronutrient intakes from the students in the present study were similar to those noted with the University of New Hampshire study population (27). Average carbohydrate intake for males was slightly lower than the previous study but higher for the females. The estimated total fat intake for males and females were higher than the totals reported for males and females by Burke et.al (27). The percentage of energy from fat for this present study is within range, however, the large standard deviations for both genders reveals a wide range of fat intakes. High variability is likely due to multiple factors. First, some subjects may have failed to consistently report added fat compared to other subjects which is one of the common sources of error in DR (39). Second, there are a large variety of food choices available throughout the UCONN campus. University Dining Services do provide healthy options or alternatives to many of the food items available daily, however, not every student consumes these items daily. These factors could result in higher variability in recorded fat intake among the students. In addition, protein intakes for males and females were slightly higher than that of the University of New Hampshire population (27). Overall, the males and females in the present study had similar macronutrient compositions in daily diet compared to previous studies among college students.

The mean daily intake for  $\alpha$ -tocopherol in the present study population was 21.3 mg/d for males and 10.2 mg/d for females. This was considerably more than reported for the U.S. nutrient intake data from NHANES 1999-2002 (104). Even lower  $\alpha$ -tocopherol intakes ( $4.6 \pm 1.6$  mg/d for males and  $4.9 \pm 1.4$

mg/d for females) were reported from a study in healthy young adults from U.S. population (164). While the sample size was larger, it was more comparative and similar age range. As stated in previously, the higher intakes and between subject variability, specifically in the male subjects, was the result of large and frequent consumption of fortified breakfast cereals from this population group.

The average vitamin C intakes for males and females in this study were 1.9 and 2.0 times higher than the EAR, respectively. Similar results were found among a healthy adult population in Europe (165).  $\beta$ -carotene intakes for males and for females was similar to Greek adults in a European study (106) that reported male and female intakes at 4532  $\mu\text{g/d}$  and 4828  $\mu\text{g/d}$ , respectively, despite larger age range and sample size. We found the variation in consumption of total flavonoids to be quite large. Chun et. al. reported a mean intake for men as  $214.1 \pm 13.8 \text{ mg/d}$  which is similar to the males in the present study (104). However, Chun et. al. reported female intake at  $200.2 \pm 12.1 \text{ mg/d}$  which is greater when compared to the female mean intake in the present study (104). The total isoflavone and total proanthocyanidin intakes in the present study were considerably higher than the intakes among the Greek male and female population who consumed  $<0.1 \text{ mg/d}$  of total isoflavone for both genders and 67 mg/d and 89 mg/d for female and males, respectively, of total proanthocyanidin (106). Estimating usual antioxidant intakes can help to establish a relationship between dietary intake and disease risk factors associated with many chronic diseases (82).

### *5.1.3. Within- and Between-Person Variation in Nutrient Intakes of Male and Female College Students*

Overall, in this study, macronutrients were more stable day-to-day in the diet than micronutrients. The participants in the 90<sup>th</sup> and the 10<sup>th</sup> percentile represent extreme intakes for each nutrient. While the 90<sup>th</sup> percentile consumed a greater amount than the 10<sup>th</sup> percentile for fat, it was to a lesser extent than with flavonoid intake. This is consistent with the results from Willett et al. when comparing fat intake to vitamin A intake (39). In studies that estimate average nutrient intake, it is usually a main objective to estimate each individual's truest intake over a long period of time. Individual intakes can, in sum, represent the entire population. Estimation of usual intake is dependent on the number of days recorded; however long term diet records are rarely employed due to participant responsibility. The effect of selecting fewer days has a greater impact for the micronutrients than for macronutrients with the overall distribution improving with the inclusion of more days of dietary assessment. Results from the present study are consistent with Willett et. al. for comparison of macronutrient to micronutrient intakes (39). These graphs provide the foundation for determining the variation for this population. In addition, they provide data to indicate that, while macronutrients do have greater stability in daily diet than micronutrients, distributions of average intake for this population require more than one day and possibly more than 7 days to become more normalized.

The within-person variation was larger than the between-person variation for most nutrients for the population in this present study. These findings are

similar to previous studies (47, 97, 99, 166). Only protein and a few micronutrients (riboflavin, vitamin B<sub>6</sub>, and magnesium) had variance ratios <1 indicating that the between-person variation was larger than the within-person variation even after adjusting for gender. The macronutrients, carbohydrate and protein intakes as well as select micronutrients such as vitamin A, lycopene, total carotenoids, B vitamins (excluding thiamin and vitamin B<sub>12</sub>), vitamin D, calcium, iron, magnesium,  $\alpha$ -tocopherol, total flavonoids, and total proanthocyanidins did not have as high of variance ratios for total participants suggesting that the day-to-day intake is more stable while the between-person variation is greater for those nutrients.

For the majority of the nutrients in this present study, adjusting for gender and energy intake did not decrease the variance ratios indicating two possible outcomes: 1) only the between-person variation was decreased which increases the variance ratios, or 2) the within-and between-person variation were decreased with the same magnitude. Overall, the within-person variation for this college population was greater than the between-person variation but only markedly different for nutrients consumed less frequently such as the carotenoids, different forms of vitamin E, vitamin B<sub>12</sub>, and isoflavones indicating the stability of the remaining nutrients in usual diet and the homogeneity of this population group as a whole.

The within- and between-person variation for the population in this study differs greatly among genders. Typically as the total intake increases, the within-person variation increases which could result in the higher coefficient of variation

within for the males in this study. While the males tended to have higher coefficients of variation, the differences between the genders for the within-person variation were small. This indicates that even though the diet composition was similar, the males were consuming higher intakes. However, the coefficients of within-person variation for various forms of vitamin E,  $\beta$ -cryptoxanthin, and lutein + zeaxanthin among the males were markedly higher than the females which suggests that the males had a more variable consumption pattern as well as a wider variety of foods containing these nutrients. The same pattern was true for the females for lycopene and isoflavones. One interesting finding was that despite the significantly higher intake of flavonoids among the males compared to the females, the females had a higher coefficient of within-person variation. One suggested interpretation of this may be that while males are consuming far more total flavonoids, the females had greater variability day-to-day and consuming a wider variety of flavonoid containing foods.

The major difference between genders was the small between-person variation among the females. This resulted in higher variance ratios among the females despite their lower within-person variation coefficients. Small between-person variation can be a result of a homogenous population which is the case in this present study. Age, BMI, and education level tend to be major determinants of a greater variation between participants in many populations (47), however, this present study consists of only young adults gaining higher education. In addition, small between-person implies less accuracy in ranking nutrient intakes

for a given study period and can increase the number of days required (47, 83). In addition, it can limit the application of results to other population groups.

In a study comparing variation of intakes among older adolescents from Russia and U.S. (99), Russian females had higher variance ratios than males which is similar to the results found in this present study; however, U.S. males had higher ratios than females. In the present study, the females had higher variance ratios for all macronutrients, iron, thiamin, niacin, and vitamin C compared to Russian and U.S. females which could be the result of difference sample sizes and slightly younger age group. However, the variance ratios for the older adolescent males and females from the U.S. population for calcium, magnesium, and riboflavin were similar with our present study. In addition, the males and females in this present study had similar day-to-day variability to their younger U.S. counterparts, which were higher than the Russian adolescents (99). In comparison to males and females  $\geq 18$  years in a previous study, the variance ratios for the macronutrients were similar to the males in this present study but slightly lower than the females in this present study (47). However, the variance ratios for the micronutrients reported by Nelson et.al. were similar to this present study for both genders but the variance ratios for vitamin A and  $\beta$ -carotene from this present study were much lower for both males and females.

The high day-to-day variability of vitamin B<sub>12</sub> for males and females was an interesting result compared to the other B vitamins. From further analysis, consumption of vitamin B<sub>12</sub> was dependent on special food items prepared at the dining halls on campus. Food sources such as the eggs in omelets and shellfish



were unique items featured on the menus on various days throughout the month of dietary assessment and accounted for the high within-person variation for both genders. However, when compared to the male and female adults reported by Nelson et.al., the variance ratios for vitamin B<sub>12</sub> in this present study was smaller for both genders (47). In addition, individual carotenoids and the polyphenols had high within-person coefficients of variation that were greater than 100. This supports the hypothesis that the results are due to the wide variety of food available on this college campus.

Overall, adjusting for energy reduced the within- and between-person variation for the macronutrients and major micronutrients for both males and females in this population group, however, did not considerably alter the variation for the carotenoids, vitamin C, or the polyphenols. It is important to note that the magnitude of the reduction in the coefficients of variation was similar between genders indicating that adjusting for energy did not considerably weaken the differences between genders. One important implication from these variance component estimates is that in order to estimate usual nutrient intakes from smaller number of dietary assessments, the mean nutrient intakes must be adjusted for the within- and between-person variation; this present study provides detailed estimates for intake of energy, macro- and micronutrients intakes as well as antioxidants for young adults.

In conclusion, energy intake accounted for the differences in intakes for most nutrients; however, quantity and frequency of consumption were responsible for the differences for  $\alpha$ -tocopherol and flavonoids between genders.

Between-person variation was smaller among females than males as evidenced by the larger variance ratios. Overall, similar diet composition was found among the genders and large coefficients of variation reflect the wide variety of food choices available to this population. Our estimation of the variance components among a population are initial step in order to determine the accurate number of days to assess usual nutrient intakes (43, 47, 114)

## **5.2. The Number of Days to Accurately Assess Usual Intakes for Energy And Key Nutrients, Including Antioxidants, among College Students**

The major finding from calculating the number of days required for accurate estimation of intakes for the total population group in this present study is, after adjusting for energy and gender, macronutrients and major micronutrients, including riboflavin, niacin, vitamin B<sub>6</sub>, folate, vitamin D, calcium, iron, and magnesium could be estimated within a 7-day period. In addition, despite the hypothesis that all antioxidants would require more days than macronutrients, lycopene and total proanthocyanidins could be estimated within 7 days and  $\alpha$ -tocopherol, total carotenoids, and total flavonoids required only 8 days for this population group. Energy, the remaining micronutrients and antioxidants for the total population would require two weeks of dietary records or more but less than one month (excluding  $\beta$ -cryptoxanthin).

In addition, there were differences between genders in estimating the number of days with females requiring more days than males for most nutrients. This is consistent with findings from Nelson et. al. from the adults population in their study (47). However, important findings in this present study pertaining to

the females were that for the nutrients thiamin, vitamin D,  $\gamma$ -tocopherol, zinc, lutein + zeaxanthin, and flavonoids required less days to assess intakes than the males. More days were required in this present study to estimate energy, fat, carbohydrate, vitamin C, and vitamin E intakes compared to the population reported by Nelson et.al. However, the age range and population size for Nelson et. al. is much larger and may contribute to their lower calculated number of days (47). Overall, usual nutrient intakes for both males and females in this population could be assessed within 30 days of dietary assessment (excluding  $\beta$ -cryptoxanthin for both genders, lutein + zeaxanthin and  $\gamma$ -tocopherol among males, and lycopene among females).

This present study is the first to provide data on the calculation of days required for accurate estimation of antioxidant intakes for this age group. Therefore, there are no direct comparisons of results to previous research. From these findings, the commonly used 1-7 day DR would not be sufficient for a precise estimate of usual nutrient intakes that includes antioxidants in this subset population of young U.S. adults. Studies intending to measure antioxidant intakes should consider the day-to-day variability in food choice and should increase the number of dietary assessments included in the study design.

### **5.3. Assessment of Nutrient Intake Adequacy and Impact of Supplement Use on Nutritional Status of College Students**

For many individuals, this transition period of young adulthood includes establishing a sense of independence and this often involves making lifestyle

decisions that impact their overall health and well being (123). Poor dietary intake and quality in conjunction with lack of regular physical activity are important contributors to the increase of health disparities that has affected every age group in the U.S. population (119, 124). However, major findings from this objective indicate that this sample population of college students participate in regular physical activity and consume adequate intakes for most nutrients.

#### *5.3.1. Lifestyle factors among college students*

A recent study identified factors that functioned as enablers or barriers to health in college students (123). They reported that being physically active, regulating food intake, social support, healthy dining options at University dining services, and University environment to support physical activity as enablers to healthful behaviors. Barriers included high stress, time constraint, monetary cost of healthy foods, ready access to fast foods, and certain social situations (123). The majority of the present study participants reported eating meals on campus in both genders and also included healthy meal options in the DR from the dining services. In a cross cultural study that analyzed physical activity levels, 58% of normal weight U.S. young adults participated in vigorous activity regularly and reported more males than females were vigorously active (125). These findings are concurrent with the males in the present study regarding vigorous activity. The selection criteria of healthy individuals for this study may explain the percentage of physical activity as well as the unique privilege of on campus gyms and many recreational sport activities available year round to students which can be considered an enabler to health.

While the college environment often facilitates great social opportunities and support systems, it can also be a time of body transitions and weight concerns, especially for women (126-128). More young adult females than males tend to diet, avoid certain foods, have lower self esteem, skip meals to lose weight, and have body dissatisfaction (95). The female participants reported similar body weight concerns trends. Over 60% of females reported that they felt social pressure to be a certain weight. In concurrence, more females than males reported a fear of gaining weight to be over a moderate amount. However, only 30% of the females reported that these body image concerns lead them to skip meals in order to lose weight. Again, the exclusion criteria for a healthy BMI range may have affected the results of these weight concerns.

#### *5.3.2. Nutrient Intake Adequacy According to the EAR Cut-Point Method*

Unlike many studies that employ only a few days of dietary assessment, the use of multiple recalls minimizes the day-to-day variability which is a major limitation in assessing usual intake for populations (40). Results from the calculation of days indicates that for the nutrients with a specific DRI, usual intakes for this population of males and females can be accurately estimated within the 30 days of dietary assessment that was collected in this present study. However, the use of the EAR cut point method requires that the nutrient intake distribution be normal (45, 158). Therefore, for this objective, the mean intakes were log transformed and back transformed to original scale. The results from this method were then used to determine nutrient intake adequacy according to the DRIs.

Results from the usual nutrient intake data indicate that students are consuming macronutrient dense foods but lower levels of micronutrients. Suboptimal dietary intakes of micronutrients have been reported to be consumed below the recommended amounts among adolescents and young adults in the U.S. population when compared to macronutrient intakes (143, 167). Results were similar among this study population of healthy, young adults for select micronutrients. A large percentage of the males and females had usual intakes below the EAR for vitamin E. For individuals, the goal for adequate intake is at an RDA of 15 mg  $\alpha$ -tocopherol per day and does not include other forms of vitamin E (59). It has been suggested that this intake level may be too high due to the fact that the usual American diet does not contain this amount (168). Results from the 1994-1996 CSFII among adults indicate that only 8% of men and 2.4% of women over the age of 20 y met the EAR. Furthermore, only 9% of men and 2.6% of women in the northern region of the U.S. met the EAR (169). In an additional analysis of the CSFII in older adolescents, 99% of females and 84% of males were below the EAR (122). A diet that is high in fruits and vegetables but lower in fat intake and processed grains would be following the Dietary Guidelines (170), however, may not be rich in vitamin E food sources (171). As with many micronutrients, clinically defined vitamin E deficiency is not common among this age group in the United States and therefore, more research is needed to explain this gap between low intake adequacies and low incidence of deficiencies among population groups (20, 171).

In 2010, the IOM released new DRIs for calcium and vitamin D (172). In previous studies, the AI was used to for vitamin D and calcium to assess nutrient intake adequacy among population groups (88, 121). However, the consensus report indicate that a new EAR of 10 µg/d and RDA of 15 µg/d for vitamin D for all life stage groups has been established and the EAR for calcium is 1,100 mg for males and females 14-18 y and 800 mg for males and females 19-30 y (49, 172). The majority of the participants in this study (76% males and 96% females) were below the EAR for vitamin D. Calcium intakes among the females were low with 41% below the EAR but only 29% of the males were below the EAR. Taking a closer look at the daily servings of dairy among this population, females reported an average of only 2.4 servings of dairy while males reported only 2.6 servings per day which the dietary guidelines suggests 2-3 servings daily (170). In comparison, another subset population of college females reported a mean intake of only 1.3 daily servings of dairy (26). While the mean servings of this present study population was within the guidelines, it was not sufficient to meet the DRI for vitamin D and calcium for many students. Similar results were found with magnesium (41% of females and 24% of males). Low intakes of vitamin D, calcium, and magnesium, specifically for females, increases their risk of poor bone health in the future (144).

Few of the students in the present study consumed inadequate amounts for vitamins A (24% of males and 8% of females) and C (12% of males and 4% of females), and folate (0% of males and 4% of females) which have a higher prevalence of under-consumption among many U.S. young and older adults

(101, 121, 122, 170). A main cause of low intakes of these nutrients is believed to be that few individuals meet the requirement for daily servings of a variety of fruits and vegetables (170). The Dietary Guidelines for Americans 2010 reports that the average U.S. adult consumed only 1.6 servings of vegetables and 1.0 servings of fruit daily and are at 59% of the goal for vegetable intake and 42% for fruit intake (170). Upon closer analysis, the males were consuming on average 2.2 servings of fruit and fruit juices and 4.5 servings of vegetables and vegetable juices while the females were consuming 2.1 servings of fruit and fruit juices and 3.4 servings of vegetable and vegetable juices. These food sources are major contributors to the average intakes of micronutrients in this population.

Methods for determining inadequacy using the EAR cut-point method are appropriate and recommended for all nutrients with a defined EAR except iron (116). Iron violates the assumption that the distribution of requirements are symmetrical (114) which is not true for women in their reproductive years due to blood and iron losses during menstruation. Therefore, the distributions of iron requirements is skewed and the proportion of individuals in a group below the EAR does not necessarily reflect prevalence of inadequacy (115). It should be noted that the mean iron intakes for both males and females were high in comparison to previous studies (37, 47, 101, 122).

Without biochemical assessment, dietary information cannot be used to define a deficiency in a certain nutrient. However, dietary assessment can be used to define who maybe at risk or determine who may benefit from approved dietary supplementation (36). In conclusion, using the EAR cut-point method,



there is a high prevalence of inadequate intakes of vitamin E, vitamin D, calcium, and magnesium for the males and females in this population group. While the remaining important nutrients had fewer individuals below the requirement, it may be of importance for nutritional interventions to be focused on improving the overall dietary intakes of micronutrients for this influential population due to usual intakes at the EAR are expected to be inadequate for 50% of individuals (117).

### *5.3.3. Supplement Usage and Contribution to Nutrient Adequacy among College Students*

Overall, 39% of the participants reported habitual supplement use. This is consistent with previous findings of 34% among adolescents (152). The most frequently consumed supplements among the population in this present study and previous research were multivitamin or individual vitamins/minerals supplement (34, 173). Individually, males have reported greater use of ergogenic aids such protein supplements while females more frequently consume herbal supplements related to weight loss (142). More of the males in this study reported use of ergogenic aids and non-vitamin, non-mineral supplements; however, none of the female students reported any use of weight loss supplements or herbal complexes. Female users in this study reported higher intakes of individual vitamins or minerals. It is of value to note that while previous studies with supplementation report higher prevalence among women (130), this present study reported more consistent use among males.

Supplement use has been associated among those with higher nutrient intake from dietary sources. Therefore, when analyzing total nutrient intake from

diet and supplements, the prevalence of supplement users with nutrient intake below the EAR has been shown to decrease (174). While supplementation may be warranted among those individuals with inadequate dietary intakes, research shows they are less likely to consume a supplement (130). Despite the prevalence of inadequacy among the participants in this study, those that were classified as supplement users overall had higher dietary intakes before supplementation.

Nutrient intake, when comparing average intakes between non-users and users, significantly increased with the addition of a supplement for all nutrients except vitamin A. However, the use of a supplement only improved adequacy among users compared to non-users for vitamins D and E, and magnesium. These three nutrients had a high prevalence of inadequacy among the males and females in the previous section. However, only the proportion of users from total diet below the EAR for vitamin D was significantly improved with supplement intake. Supplements containing vitamin E did greatly improve the adequacy among users but the lack of significance can possibly be attributed to a low population size. This improvement with supplementation of these vital nutrients may imply that supplement use should be included in future health interventions due to the high prevalence of inadequacy from dietary intake sources alone reported among national surveys (101). However, as stated previously, the concern is with the non-users who do not consume a supplement and had significantly lower intakes from dietary sources compared to the diet of the users. The only nutrients consumed more adequately among non-users were vitamins A

and C. While the intake of supplements containing vitamin C did improve adequacy among users, the proportion of individuals below the EAR remained greater than non-users which can be explained by the differences in age related requirements between the two groups as well as the individuals with low dietary intakes may not have taken a supplement that contained vitamin C. Supplements containing vitamin A did not account for much of the average intake as it did not significantly increase intakes between non-users and users or decrease the proportion of users below the EAR. It is of interest to note that with the increasing data of the benefits of antioxidants, many supplements now include various forms of flavonoids such as green tea supplements found in this study. Further research is implicated in the bioavailability and health benefits of supplement forms of these nutrients.

Supplements are over a 25 billion dollar industry in the United States and many remain unregulated (151). The naivety and often unadvised usage of this population group puts them at risk for adverse side effects of over supplementation (152). Most nutrients consumed at or above the UL from dietary sources only have not been shown to have adverse effects. The issue arises with supplemental forms or fortification of foods (36). While intakes of folate, niacin, iron, and zinc were consumed above the UL in this population, there were no significant differences between intakes from dietary sources only and diet + supplement intake among users. However, the use of ULs to assess risk of adverse effects has its limitations when assessing a population due to varying sensitivities among individuals (117).

Using the EAR cut-point approach for assessing total nutrient intake adequacy, we found that supplement users benefited significantly from supplement use compared to non-users resulting in more adequate dietary intakes. The effects of energy intake differences were removed; therefore, these results imply that the diets were inadequate in many micronutrient dense foods. This research provides novel data regarding long term nutrient intake, supplement contribution to meeting nutrient requirements, and lifestyle factors among a healthy, young population.

#### **5.4. Conclusion and Future Direction**

The overall aim of this thesis was to estimate usual nutrient and supplement intake that included antioxidant nutrients from long term, consecutive diet records. In addition, validity of self reported diet records were analyzed, nutrient intake adequacy from major macro- and micro-nutrients was assessed, and the number of total diet records necessary to estimate antioxidants was calculated.

This study had strengths and limitations. The first strength was the identification of misreporters in a young adult population which is vital when reporting nutrient intake data. As suggested by Black et al., this study included physical activity questions in the health and nutrition survey and therefore, defined a specific PAL for the population in this present study (40). The second strength was the utilization of 30 consecutive days of dietary assessment data in order to represent more habitual nutrient intake by reducing the day-to-day

variability. In addition, the number of days to estimate usual antioxidant intake was performed in a population group that is not frequently studied. Major limitations include: lack of seasonal variation representation, however, seasonal variation tends to have a greater effect in developing countries where food availability is more dependent on environmental factors than in the U.S. (39); the confidence limits defined by the Goldberg cut-off equation did not account for true energy expenditure among this population; intake levels reported by the participants are interpreted without any confirmation by biochemical data; finally, there may be some nutrient selection bias in the population in this study as a result of homogenous inclusion criteria pertaining to health status and education level which can limit the applicability of the results to other population groups.

In conclusion, 15% of males and 30% of the females underreported dietary intakes which is lower than previous studies and implies accuracy in reporting for this population. For the majority of nutrients, males did not consume significantly higher intakes, after adjusting for energy, compared to females. In addition, more females than males consumed inadequate nutrient intakes. Supplement users had significantly higher total nutrient intakes than non-users and therefore, had more average intakes above the guidelines. Micronutrients had greater variation when compared to macronutrient intakes. For most nutrients, females required more days of dietary records. Overall, estimation of usual intake status would require a 7-day DR or more for this well educated, healthy young adult population. There has been limited research to estimating dietary intake of flavonoids, isoflavones, and proanthocyanidins due to lack of

polyphenol food composition data. However, the present study represents necessary steps to estimate accurate nutrient intake and provides data on average antioxidant intakes in this population as the first step to establish a relationship between diet and disease.

In future studies, a larger sample size can be used to estimate usual intakes, including antioxidants, in order to establish a relationship between dietary intakes and the risk of disease among this young adult population group. In addition, more epidemiological and clinical studies need to consider misreporting nutrient intake and the number of dietary records to assess antioxidant intakes in the study design due to the large day-to-day variability. There is a need for more studies to be conducted to determine nutrient adequacy, supplementation, and lifestyle factors associated with this population. Food and nutrition professionals should address possible interventions to improve nutritional quality among the young adult population. Overall, the present research contributes data that suggests increasing the number of days of dietary assessment to estimate usual antioxidant intakes in any population group is required to create a stronger relationship between dietary intakes and disease risk factors for these important nutrients.

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